Comparing Whiteboard and 3D Animation in Visualization of Neuron-like Bacterial Communication in Biofilms

BY

SARAH MCGUINNESS
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THESIS

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Defense Committee:

Christine Young, Chair and Advisor
Dr. Andrew Boyd
Dr. Evelyn Maizels
Rex Twedt

This thesis is dedicated to my research advisor, Christine Young, without whom it would never have been accomplished.

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SUMMARY

Discovery research in Dr. Arthur Prindle's laboratory has revealed a novel form of bacterial communication through ion channel-mediated electrochemical signaling (Lee, Prindle, Liu, & Süel, 2017) resembling neuronal signaling (Pan et al., 2008 ; Petroff, Errante, Rothman, Kim, & Spencer, 2002). Because 80% of chronic bacterial infections are biofilm-associated, coordination between scientists, clinicians, and the healthcare industry is necessary to find a long-term solution for bacterial biofilms (Donlan, 2008). The most effective way to deal with bacterial biofilms is to understand and disturb their physiology by interrupting their communication (Anderson & O'Toole, 2008).

Animations have become increasingly predominant in their use of teaching spatial and temporal relationships and complex processes (Ploetzner & Lowe, 2012). Unfortunately, animation is not universally successful as a learning tool, and it risks causing cognitive overload via increasing extraneous processing beyond working memory's limitations (Betrancourt & Tversky, 2000 ; Sweller, 2005). Additionally, computer-based animation is expensive and can require significant technical expertise (Mayer & Jackson, 2005).

Whiteboard animation can be a useful and efficient tool for communicating lesser-known, complex concepts (such as bacterial electrochemical signaling) to health professionals. The whiteboard represents a unifying experience for users: it's familiar, comfortable, and appealing (Wears, Perry, Wilson, Galliers, & Fone, 2007). Harnessing this emotional appeal through whiteboard-style animation may be critical for animation-based learning. However, the academic research on whiteboard animation is limited, and the research has yet to define best practices for the development of a whiteboard-style animation. Whiteboard animation has many innate advantages which may allow it to bridge the gap between static graphics and animations. A common component of whiteboard animation, the presence of a drawing hand has a significant effect on cognition (Castro-Alonso, Ayres, & Paas, 2014). Observing an instructor's hand drawing diagrams promotes the learning process by exploiting the principles of multimedia learning (Fiorella & Mayer, 2016).

The results of the survey indicated that although 58% of participants responded that the whiteboard animation was easiest to understand, the preference for learning tool was evenly divided between whiteboard and 3D animation. Furthermore, most participants enjoyed the 3D animation more, indicated that the 3D animation is more successful at communicating complex information, are more likely to recommend the 3D animation to peers. All knowledge transfer scores showed an increase in knowledge across pre- and post-tests. More data will be obtained and analyzed to determine if there is a statistically significant difference in knowledge transfer scores between participants who viewed the whiteboard animation first and those who viewed the 3D animation first.

This project's results may inform best practices for whiteboard animation relying on the conceptual frameworks of human cognition and multimedia learning. This project may also lead to further applications of whiteboard animation in biocommunication between health professionals and scientists. Whiteboard animation provides a more accessible threshold animation in that it can be less demanding on time, resources, money, and technical expertise, and it can potentially avoid cognitive overload, providing a highly efficient mechanism of biocommunication.

I. INTRODUCTION

A. Overview of research problem

Animations have recently become increasingly predominant in their use of teaching four dimensional relationships and complex processes (Ploetzner & Lowe, 2012), especially in teaching (Hoyek, Collet, Di Rienzo, De Almeida, & Guillot, 2014 ; Singh, Singh, & Gautam, 2009) and molecular and cell biology education (McClean et al., 2005 ; O'Day, 2006). Unfortunately, animation is not universally successful as a learning tool, and it risks causing cognitive overload via increasing extraneous processing beyond the limits of working memory (Betrancourt & Tversky, 2000 ; Sweller, 2005). Much of the research surrounding the efficacy of animation as an instructional tool is inconsistent and contradictory, due to methodological issues (Betrancourt, 2005 ; Mayer, Hegarty, Mayer, & Campbell, 2005). Some studies have even found animation to be detrimental to learning (Betrancourt, 2005). Additionally, computer-based animation is expensive and requires technical expertise (Mayer & Jackson, 2005). For subject matter high in complexity, instructional design must rely on cognitive load theory to develop a tool with legitimate efficacy (Sweller, 2005). Unfortunately, instructional design does not always follow the principles constructed from cognitive load theory and multimedia principles (Sweller, 2005).

A newly discovered method of cell-to-cell bacterial biofilm communication mediated by potassium ion channels, creating neuron-like long range cell signaling (Prindle et al., 2015), presents a significant communication challenge in that it is inherently complex and unique. In addition, the audience has limited knowledge of biofilms or neuronal communication. Visualizing, comprehending, and understanding even fundamental molecular or cellular processes can be very challenging for college students (McClean et al., 2005). For audiences in higher education or health professions, such as graduate, medical, nursing, or dental students, this research seeks to develop the most efficient tool to communicate a complex and interdisciplinary topic. The educational challenge involves both a topic that is little known and inherently complex. In addition, student populations are evolving their learning styles and visual preferences, as the digital generation transitions into adulthood. A recent study done with medical students showed that their higher affinity for information communication technology experienced a disadvantage in lecture-based classroom environments (Backhaus, Huth, Entwistle, Homayounfar, & Koenig, 2019). Now, information in the form of images, sound, text, and video can be transmitted instantly (Ayres & Williams, 2004). To accommodate this transformation in communication and the advancements in learning theory over the past decade, new teaching methods must be developed which account for these evolving preferences.

B. Significance of the problem

Biofilms may be responsible for chronic bacterial infections (Bjarnsholt, 2013) and may be resistant to antibodies, phagocytes, and antibiotics (Berk et al., 2012). Their architecture allows them to proliferate in environmental, medical, and industrial systems (Lopez, Vlamakis, & Kolter, 2010b ; Spormann, 2008). In the hospital environment, biofilms can adhere to surgical site sutures and cause severe morbidity and mortality, resulting in an annual cost of $1.5 billion in the US (Seal & Paul-Cheadle, 2004). Over 17 million cases of biofilm associated infections occur every year in the United States, resulting in over 500,000 deaths and an annual cost of $94 billion (Wolcott RD et al., 2010). Even the usage of antimicrobial agents is limited, because of the risk of toxicity to patients and the possibility of antimicrobial resistance (Donlan, 2008). Coordination among scientists, clinicians, and the healthcare industry is necessary to find a long-term solution to lessen the medical impact of microbial biofilms (Donlan, 2008). The creation of a visual teaching tool that could efficiently elucidate this novel method of biofilm communication could aid the scientific community in developing and communicating future strategies to manage biofilm resilience. The educational challenge is the optimization of developing informational technologies for a student population that has grown up in an informational society, although teachers primarily use traditional methods and tools (Mata, Lazar, & Lazar, 2016). The whiteboard represents a unifying experience for users: it's familiar, comfortable, and appealing (Wears et al., 2007). Harnessing this emotional appeal through whiteboard-style animation may be critical for animation-based learning. However, the academic research on whiteboard animation is limited, and the research has yet to define best practices for the development of a whiteboard-style animation.

II. LITERATURE REVIEW

A. Biofilm Formation and Growth

Escherichia coli, Bacillus subtilis, Staphylococcus aureus, and *Pseudomonas aeruginosa* are commonly studied in laboratory settings for biofilm growth (Lopez et al., 2010b). The Gram-positive *B. subtilis,* a sporulating rod-shaped bacterium with the potential for motility, is a traditional model organism for observing biofilm development (Grossman, 1995). All bacteria can form biofilms, which are architecturally complex multicellular communities connected via an extracellular matrix, allowing multiple bacterial organism to function as one (López & Kolter, 2010 ; Lopez et al., 2010b). Even some archaea and eukaryotes, such as fungi, can form biofilms (Hall-Stoodley, Costerton, & Stoodley, 2004). The process of biofilm formation is a tightly regulated transition from a motile unicellular state to a nonmotile multicellular state (Lemon, Earl, Vlamakis, Aguilar, & Kolter, 2008). Specialized cell types differentiate as a function of environment, creating a phenotypically heterogenous multicellular society, within which phenotypically distinct subpopulations exist (Costerton, Stewart, & Greenberg, 1999 ; Hall-Stoodley et al., 2004 ; Lopez et al., 2010b).

The biofilm architecture is a defense mechanism for bacterial communities to survive hostile, high stress environments (Berk et al., 2012 ; Costerton et al., 1999 ; Hall-Stoodley et al., 2004). Biofilm development is affected by many factors, including temperature, pH, oxygen levels, fluid movement, osmolarity, the presence of specific ions, nutrients, and other microbes (Goller & Romeo, 2008). The source of this environmental stress can be either external or self-induced, like competition for resources (Spormann, 2008). As a result, microenvironments within the biofilm develop and cause both physiological and genetic changes in the biofilm's subpopulations (Spormann, 2008). The nature of growth-limiting nutrient, such as an electron donor or acceptor, phosphorus, oxygen, or nitrogen, and the diffusion of substrates into and metabolites out of a biofilm are responsible for distribution of chemical gradients throughout the biofilm (Spormann, 2008).

Biofilm formation can be separated into five discrete, tightly regulated, stages: surface attachment, the formation of a monolayer, migration, secretion of an extracellular matrix, and the maturation of the biofilm with characteristic three-dimensional architecture (Lemon et al., 2008). Motile bacteria, like *B. subtilis,* can transition from a planktonic state to a nonmotile state through a complex and highly regulated process (Lemon et al., 2008 ; O'Toole, Kaplan, & Kolter, 2000). In the first stage of biofilm formation, an external stress signal induces the bacterial cell to attach to a surface (de la Fuente-Nunez, Reffuveille, Fernandez, & Hancock, 2013). Individual cells can increase their stickiness by expressing more adhesins on their outer surface (Lemon et al., 2008). Biofilms prefer to transition between an aqueous environment and a solid substrate, or between a gaseous phase and a solid substrate (Spormann, 2008). This survival feat is made possible by the extra cellular matrix of exopolymeric substance (EPS) surrounding the biofilm, created by the resident bacteria (López & Kolter, 2010a ; Lopez et al., 2010b). This complex extracellular matrix allows for cell-cell adhesion, adhesion to a surface, and formation of a protective yet flexible envelope which encases the biofilm (Berk et al., 2012). The final stage of biofilm formation is proliferation of the bacterial community in a complex three-dimensional architecture, complete with extracellular signaling and entirely encased in a protective layer of EPS (de la Fuente-Nunez et al., 2013 ; Lemon et al., 2008 ; O'Toole et al., 2000). After a biofilm has matured, it can initiate designated dispersal (De

Koning, Tabbers, Rikers, & Paas, 2009). Some cells dissociate from the biofilm in order to colonize new surfaces or flee the biofilm's environmental limitations on bacterial growth (Goller & Romeo, 2008 ; O'Toole et al., 2000). The biofilm's matrix can prevent egress of its members, but environmental changes, such as changes in specific nutrients, can trigger large-scale dispersal by inducing genetic expression of motility (Goller & Romeo, 2008).

These highly adaptive heterogenous microbial societies are difficult to treat with current antimicrobial solutions (Costerton et al., 1999 ; Spormann, 2008). Biofilms combine both innate and induced mechanisms for antibiotic resistance, ultimately resulting in a highly recalcitrant microbial community and increasing antibiotic resistance 10- to 1,000-fold (Anderson & O'Toole, 2008). High temperatures, UV radiation, oxidants, disinfectants, and antimicrobial coatings can be used to combat and prevent biofilms on surfaces, such as metal or plastic, (de Carvalho, 2007), but fighting bacterial biofilm infection in the human body requires alternative methods. Antibodies, phagocytes, and antibiotics have limited efficacy against microbial biofilms (Costerton et al., 1999), and an external attack may even promote growth and viability within the biofilm (Liu et al., 2015). Although antimicrobial agents can be used to eliminate bacterial biofilms, either systemically or via an antimicrobial lock treatment, their ability to do so is limited and the use of these agents at high concentrations can cause toxicity in the patient (Donlan, 2008). Even more concerning, the potential of antimicrobial resistance, especially in the most vulnerable of patient populations, demonstrates the need to limit antimicrobial agent usage (Donlan, 2008).

Biofilms can be found on a rich variety of surfaces. They can invade most environmentally, medically, and industrial relevant systems (Spormann, 2008), even highly irradiated nuclear power plants (de Carvalho, 2007). Biofilms can be responsible for painful disease or infection, but they can also wreak havoc on ship hulls, tubes, and pipes (de Carvalho, 2007). They can cause infections on human teeth, the oral cavity, gingival crevices, skin, urinary tracts, medical devices and implants, and reside in intestinal mucosa, and even the lungs of cystic fibrosis patients (de Carvalho, 2007 ; Eckburg et al., 2005 ; Goller & Romeo, 2008 ; Hatt & Rather, 2008 ; Kolenbrander, 2000).

The adherence of bacterial biofilms to indwelling medical devices culminates in severe morbidity and mortality (Donlan, 2008). Among the medical devices vulnerable to infection are catheters, endotracheal tubes, mechanical cardiac valves, contacts, prosthetic joints, and surgical sutures (de Carvalho, 2007). Surgical site infections contribute heavily to prolonged illness and mortality, and approximately 500,000 surgical site infections occur every year (Seal & Paul-Cheadle, 2004). The National Institute of Health reports that over 60% of microbial infections and 80% of chronic infections are associated with biofilms (Jamal et al., 2018 ; Lewis, 2001). Treatment for biofilm-associated infections in America results in an annual cost of $1.5 billion (Seal & Paul-Cheadle, 2004). Although microbial biofilms remain a significant health threat, there are no known comprehensive treatments, only regimens which target specific microorganisms or biofilm formation stages (Kumar, Alam, Rani, Ehtesham, & Hasnain, 2017). Solving the biofilm problem requires coordination between scientists, infectious disease clinicians, and the healthcare industry (Donlan, 2008).

B. <u>Biofilm Metabolism</u>

The most effective way to manage biofilms is to understand and disturb their physiology by interrupting their communication (Anderson & O'Toole, 2008). Communication is vital for a community of genetically distinct organisms and, until recently, there was limited knowledge of metabolic communication within biofilms (Kolenbrander, 2000). With initial surface attachment and monolayer formation, bacteria switch their lifestyle from a nomadic and unicellular state to a sedentary and multicellular state, characterized by structured communities and cellular differentiation, is marked by metabolic changes (Lemon et al., 2008). A mature biofilm's transcriptome becomes more of a stationary phase culture than an exponentially growing culture (Goller & Romeo, 2008). This slowed metabolism is significant because it happens to present an innate biofilm antibiotic resistance mechanism (Anderson & O'Toole, 2008). The shift to stationary metabolism helps multiple bacterial organisms and subpopulations of differentiated cells function as one by increasing the production of secondary metabolites, which are involved in signaling, biofilm formation initiation, or inhibition of competing biofilms (López & Kolter, 2010a ; Lopez et al., 2010b). But most, importantly, regulation of biofilm metabolism faces an innate challenge: the competition for nutrients between individual peripheral and interior cells (Liu et al., 2015).

B. subtilis biofilms, the subjects of bacterial biofilm communication in Dr. Arthur Prindle's laboratory at Northwestern University Medical School, have been found to demonstrate metabolic oscillations supporting their expansion and growth, despite growing in experimental conditions with constant media flow and thereby constant nutrient supply (Liu et al., 2015). These oscillations solve a conflict inherent to biofilm expansion: the conflict between the demands of biofilm growth and the protection of the vital interior cells, upon which the success of the biofilm relies (Liu et al., 2015). These metabolic oscillations are driven by the codependence between the biofilm's peripheral cells and interior cells and environmental nitrogen limitation (Gunka & Commichau, 2012). *B. subtilis* relies on glutamate as its only source of nitrogen and can be used to produce ammonium via glutamate dehydrogenase (Branda, González-Pastor, Ben-Yehuda, Losick, & Kolter, 2001 ; Gunka & Commichau, 2012). However, ammonium is the limiting factor for biofilm growth (Liu et al., 2015). Because ammonium will freely cross the bacterial cell membrane, bacterial cells prefer to use extracellular ammonium (Boogerd et al., 2011 ; Castorph & Kleiner, 1984 ; Jayakumar, Schulman, MacNeil, & Barnes Jr., 1986 ; Kim et al., 2012 ; Kleiner, 1985). Peripheral cells consume glutamate during growth, which starves the interior cells, yet they rely on extracellular ammonium produced by the interior cells (Liu et al., 2015). The solution to this problem is a spatial and temporal organization of metabolic activity: the peripheral cells halt growth, allowing the interior cells to consume glutamate (Liu et al., 2015). In turn, the interior cells produce and release ammonium, which can readily diffuse into peripheral cells (Liu et al., 2015).

C. <u>Bacterial communication</u>

Quorum sensing, defined as "the regulation of gene expression in response to fluctuations in cell-population density" (Miller & Bassler, 2001, p.165), is the most well-known cell-to-cell communication involving chemical signals, called autoinducers (Miller & Bassler, 2001). These signals, which can be small peptides or acylated homoserine lactones, are produced as a function of cell population density and allow bacteria to behave as multicellular organisms (Miller & Bassler, 2001 ; Waters & Bassler, 2005). Quorum sensing can be used to coordinate various physiological processes, such as virulence, sporulation, symbiosis, competence, conjugation, and biofilm formation (Miller & Bassler, 2001). Communication via quorum sensing provides the opportunity to reprogram behaviors in biofilms and potential to develop antimicrobial therapies (Miller & Bassler, 2001; Waters & Bassler, 2005).

In biofilms, individual cells must function as a collective to ensure survival (Lopez et al., 2010b). Peripheral and interior cells compete with each other for nutrients, and peripheral cells must protect the interior from external threats (Liu et al., 2015). Electrochemical signaling allows bacterial communities to achieve this balance (Prindle et al., 2015).

There are three fundamental methods of electrochemical communication found within biofilms (Lee et al., 2017). The first is short-range signaling via direct contact, such as communication through membrane-bound cytochromes (McGlynn, Chadwick, Kempes, & Orphan, 2015). The second is passive diffusion of electrons via soluble redox-active molecules (Marsili et al., 2008), in which communities of Gram-negative and Gram-positive bacteria are capable of electron transfer between cells at a distance. The third method is long-range communication using ion channel-mediated active signaling (Lee et al., 2017). This mechanism can also be found in human neurons (Pan et al., 2008 ; Petroff et al., 2002). Unlike quorum sensing, the strength of these signals is not related to population density (Mas et al., 2015).

The purpose of these ion channel-mediated “neuron-like” signals is to communicate metabolic stress over a long distance (Prindle et al., 2015). Sparked by the stress of nitrogen limitation (Liu et al., 2015), cells release intracellular potassium (K+) in a stage of transient depolarization (Prindle et al., 2015). Adjoining cells become depolarized as extracellular potassium levels increase, which causes the electrical component of each cell’s proton motive force to decrease (Prindle et al., 2015). Proton motive force (PMF) is the electrochemical gradient of protons across a membrane and consists of a pH gradient and an electrical potential

(Krulwich, Sachs, & Padan, 2011). As a result of decreased proton motive force, cellular uptake of glutamate and retention of ammonium decreases (Boogerd et al., 2011 ; Tolner, Ubbink-Kok, Poolman, & Konings, 1995), allowing interior cells to gain access to a nitrogen source (Prindle et al., 2015).

D. **Neuron Signaling**

Long-range electrochemical communication within and between biofilms has been compared to the electrical signals transmitted by neurons (Prindle et al., 2015). A neuron consists of three functional elements: dendrites, an axon, and the soma (Gerstner & Kistler, 2002). Dendrites receive signals and transmit them to the soma, the central processing unit which generates an output signal (Gerstner & Kistler, 2002). The axon delivers this signal to propagating neurons across synapses, or junctions between neurons, as shown in Figure 1B (Gerstner & Kistler, 2002). The nature of this electrical signal is a spike or pulse with an action potential of 1-2 ms and an amplitude of 100 mV, depicted in Figure 1A, the form of which remains the same for the duration of the signal propagation (Gerstner & Kistler, 2002). Synaptic transmission involves three different types of ion channels: calcium-activated, voltage-activated, and transmitter-activated (Gerstner & Kistler, 2002). When we talk about long-range biofilm communication, we focus on the latter two types.

Potassium ions have a single positive charge (Gerstner & Kistler, 2002). In both microbial and human cells, the intracellular ion concentration of potassium exceeds the extracellular concentration (Gerstner & Kistler, 2002). Sodium ions also have a single positive charge but exist in higher concentrations outside the cell (Gerstner & Kistler, 2002). Therefore, at the resting potential of the membrane, sodium ions can readily flow into the cell and potassium ions can flow. A wave of depolarization via potassium can result in voltage activated synaptic transmission (Gerstner & Kistler, 2002).

There are structural similarities between ion channels and functional similarities between long-range electrical communication in bacteria and neurons (MacKinnon, 2004; Prindle et al., 2015). Glutamate, the only source of nitrogen for *B. subtilis* biofilms (Branda et al., 2001; Gunka & Commichau, 2012) is involved in the regulation of metabolic oscillations by triggering potassium release (Liu et al., 2015; Prindle et al., 2015) and is used in the vertebrate central nervous system as a neurotransmitter to signal to excitatory synapses (Gerstner & Kistler, 2002). Understanding and further research into these similarities may help scientists develop potential solutions to biofilm associated infections.

E. Potassium Ion Channels in Bacteria

Both Gram-negative and Gram-positive bacteria accumulate potassium, the major intracellular cation (Epstein, 2003). Potassium is necessary for cellular viability, including the viability of eukaryotic cells (Epstein, 2003). In bacteria, K+ regulates internal pH, acts as a second messenger, functions as an osmotic solute, and activates intracellular enzymes (Epstein, 2003). Potassium channels extend across the bacterial cell's membrane and create a pore through which ions can selectively diffuse (Sansom et al., 2002). YugO, a putative potassium ion channel, has been found to positively regulate biofilm formation in *B. subtilis* (Lundberg, Becker, & Choe, 2013). YugO can be activated by glutamate starvation (Prindle et al., 2015) and will behave as a K+ efflux pump to induce biofilm formation (Sansom et al., 2002).

The 3D structure for the YugO channel used for my animation was a homology model constructed from an MthK K+ channel from the *Methanobacterium thermoautotrophicum*

(Jiang, Lee, Chen, Cadene, Chait, & MacKinnon, 2002). An experimental model of YugO found on SWISS-MODEL Repository (ID:Q795M8) was displayed as a homo-8-mer and a homo-4-mer. However, the literature did not confirm either representation, although the majority of potassium ion channels are tetrameric (Pollard, Earnshaw, Lippincott-Schwartz, & Johnson, 2017), nor was YugO available on PDB.

UniProt showed that both YugO (Q795M8) and MthK(O27564) share RCK domains, which form gating rings from multiple subunits (Giraldez & Rothberg, 2017). RCK domains are also found in eukaryotic K+ channels and are highly conserved (Giraldez & Rothberg, 2017). It was found that the MthK transmembrane ion channel pore is a tetramer with four full length chains, with an octameric RCK gating ring (Giraldez & Rothberg, 2017 ; Jiang et al., 2002).

F. <u>Cognitive Load Theory</u>

Cognitive load theory presents a structure for the human cognitive architecture based on biological evolution (Sweller, Ayres, & Kalyuga, 2011), which carries heavy significance for instructional design. However, cognitive load theory is not always applied to instructional practices and may even seem counterintuitive in its application (Sweller et al., 2011). Unfortunately, the effectiveness of instructional design without an understanding of human cognitive architecture is likely arbitrary (Sweller, 2005). The genetic code is as essential to human biology as long-term memory is to human cognition (Sweller, 2003). Both human biology and cognition have been environmentally molded by the laws of natural selection and adaptation (Sweller, 2003). As time passes, new information is learned in working memory and placed within long term memory, just as new information enhances the genetic code over time (Sweller, 2003).

In order to effectively design instructional tools, the limitations of working memory must be understood (Sweller, 2005). Working memory is severely constrained by two factors: it can only hold seven discrete elements of information at a time, and it can only manipulate 2 – 4 elements (Miller, 1994). Additionally, it takes only 20 seconds for those elements to be lost (Peterson & Peterson, 1959). When a learner encounters new information, their working memory become strained because of a lack of an organizing central executive entity; however, existing schema from long-term memory storage can alleviate this strain by fulfilling this logical, organizational role as a central executive (Sweller, 2005). A schema is defined as a cognitive organizational construct which allows many elements of information to be treated as one element of information (Sweller, 2005). Schemas can be brought from long-term memory into working memory in order to integrate new information (Sweller, 2005). It has been shown that continuous human perception is automatically segmented into nested or discrete events, creating the foundation for long-term memory (Baldassano et al., 2017). However, when the learner encounters alien material, they may not have pre-existing schema upon which they can build from (Sweller, 2005). When new information is taught, the instructional design must satisfy the role of the missing central executive in information integration by providing a schema for the new information (Sweller, 2005). This way, the learner's working memory can focus on using its resources for the construction and automation of schema (Sweller, 2005).

Cognitive load theory defines three distinct categories of cognitive load: intrinsic cognitive load, extraneous cognitive load, and germane cognitive load, all of which are determined by element interactivity (Sweller, 2005; Sweller, 2010). An element refers to a discrete unit of information (Sweller, 2011). Intrinsic cognitive load refers to the inherent complexity of the novel information (Sweller, 2005). For example, quantum physics has a higher intrinsic cognitive load than Newtonian mechanics. Extraneous cognitive load results when instructional design does not acknowledge the ways in which working memory is limited (Sweller, 2005). For example, giving a physics lecture which forces the learner to hold more than seven elements in their working memory at a time increases extraneous cognitive load. Germane cognitive load is functional cognitive load: it is involved in the creation of schema (Sweller, 2005). If a Newtonian mechanics class is taught by giving an example of the trajectory of a bouncing ball to illustrate how the ball's acceleration, velocity, and height change over time, the learner's germane cognitive load increases but allows them to create a schema in their long-term memory for holding this new information. Extraneous, intrinsic, and germane cognitive load are cumulative properties, the sum of which has a finite limitation due to the characteristics of working memory (Sweller, 2005). Therefore, instructional design can afford to be poorly done when the material being taught is relatively simple (Sweller, 2005). However, when subject complexity is high, instructional design must work to reduce extraneous cognitive load, manage intrinsic cognitive load, and maximize germane cognitive load (Sweller, 2005).

G. <u>Principles of Multimedia Learning</u>

When designing multimedia learning tools, it is essential to maintain the reciprocation of theory and practice, address authentic learning situations, and test learning theory (Mayer, 2008). Multimedia learning, or learning from both words and images (Mayer, 2008), may include learning from power point presentations, animations and videos, illustrations, and motion media (Figure 2).

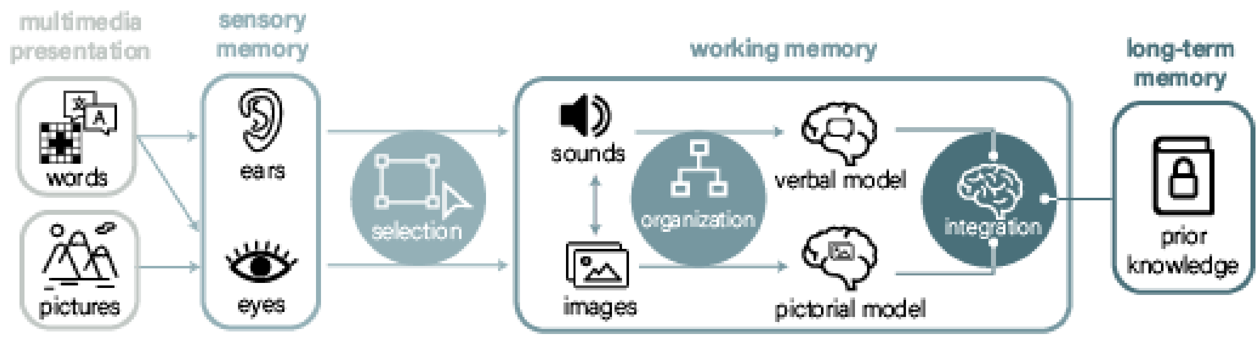


Figure 1. Cognitive Theory of Multimedia Learning after Mayer (Mayer, 2008). Graphic generated from composite of icons from the Noun Project: "Brain" by Meaghan Hendricks is licensed under CC; "Crossword" by Zach Bogart is licensed under CC; "Ear" by Ben Davis is licensed under CC; "Eye" by Antonio Herrera, ES is licensed under CC; "lock book" by Travis Avery is licensed under CC; "pictures" by lastspark is licensed under CC; "Mountains" by Flatart is licensed under CC; "selection" by Hea Poh Lin is licensed under CC; "selection" by DesignBite is licensed under CC; "Sound" by Dávid Gladiŝ is licensed under CC; "Speech bubble" by Alfredo @IconsAlfredo.com is licensed under CC; "translate" by Ivan Kostriukov is licensed under CC

A learner's cognitive process can be organized into three consecutive steps: *selecting* the input from words and pictures, then *organizing* the new information into verbal and pictorial models, and finally *integrating* these models into long-term memory by forming connections with prior knowledge (Mayer, 2008 ; Mayer, Heiser, & Lonn, 2001). There are three fundamental elements defined in the science of learning: dual channels, limited capacity, and active processing (Mayer, 2008). *Dual channels* refer to the separate neural pathways which process visual and verbal information (Mayer, 2008). *Limited capacity* acknowledges that each of these pathways is limited in how much information it can process at one time (Mayer, 2008). *Active processing* indicates that knowledge acquisition is dependent on cognitive processing during learning—selection, organization, and integration (Mayer, 2008; Mayer et al., 2001).

A multimedia learning tool must be designed to minimize extraneous processing, manage essential processing, and encourage generative processing (Mayer & Jackson, 2005). Extraneous processing, brought about by ineffective and disorienting instructional design, is defined as unnecessary cognitive processing which does not relate to the ultimate instructional goal (Mayer, 2008). There are five different principles involved in the reduction of extraneous processing: the coherence principle, the signaling principle, the redundancy principle, the spatial contiguity principle, and the temporal contiguity principle (Mayer, 2008). According to the *coherence principle,* people retain information more efficiently when irrelevant elements are removed (Mayer, 2008). Under the *signaling principle,* the most important concepts in the lesson are emphasized, either verbally or visually (Mayer, 2008). This principle is especially effective for individuals who have a limited prior knowledge of the subject matter (Richter, Scheiter, & Eitel, 2016). Some of the techniques used may include highlighting certain words/phrases, including overview sentences before a lesson, or adding section headings (Mayer, 2008). The *redundancy principle* dictates that learners acquire knowledge more effectively from animation and narration than they do from a narrated animation that includes text on the screen (Mayer, 2008). The principle of *spatial contiguity* asserts that interrelated images and text must be near to each other, whereas the principle of *temporal contiguity* argues that interrelated narration and animation must be presented simultaneously (Mayer, 2008).

However, reducing extraneous processing may not be enough. Essential processing is cognitive labor required to hold new information in working memory (Mayer, 2008). A lesson high in complexity, such as physics or microbiology, may contain so many elements and relationships between these elements that the learner becomes overwhelmed due to the high demand of essential processing (Mayer, 2008). To regulate these demands, an instructor may utilize the principles of segmenting, pretraining, and modality (Mayer, 2008). *Segmenting* avoids a continuous uninterrupted presentation and divides the information into discrete, digestible chunks, while allowing the learner to choose to move forward upon consuming each segment (Mayer, 2008). For lessons involving an overwhelming number of elements, the *pretraining principle* introduces and characterizes each element individually, then presents them together in a system (Mayer, 2008). To avoid the trap of split attention (resulting from integrating both images and written text), instructors should employ the *modality principle* and present words as auditory narration (Mayer, 2008).

Finally, stimulating generative processing, which involves understanding the new material and integrating it into networks of prior knowledge, can be induced by *the multimedia principle* and the *personalization principle* (Mayer, 2008). The former states that presenting both words and images (as opposed to words alone) promotes deep learning (Mayer, 2008). The latter suggests employing a conversation style of narration or text is more effective than a formal style (Mayer, 2008).

H. Animation as a Scientific Learning Tool

Multimedia learning has been shown to significantly increasing learning retention and student engagement (McClean et al., 2005) when applied to molecular and cell biology. However, factors like prior knowledge and context, subject complexity, and topic can all influence the effectiveness of a multimedia learning tool (Lowe & Boucheix, 2011). It is a priority that these interrelationships be elucidated for multimedia learning resources to be most efficiently designed.

It has been shown via meta-analysis that animations have an overall positive effect on learning, as opposed to static graphics (Berney & Betrancourt, 2016). However, this success is not a universal benefit, since there are several factors in animation which moderate its success (Berney & Betrancourt, 2016). Research intended to illuminate the efficacy of animation as a multimedia learning tool is highly variable due to the animation's intended purpose, the study's learning objectives, and the comparison groups (Betrancourt & Tversky, 2000). Under specific conditions, animation can positively impact both learning performance and attitude, although one must be aware that the tool itself cause cognitive overload (Betrancourt & Tversky, 2000).

Animation can be defined as "any application which generates a series of frames, so that each frame appears as an alteration of the previous one, and where the sequence of frames is determined either by the designer or the user" (Betrancourt & Tversky, 2000, p. 160). The unique strengths of an animation are the media's ability portray spatial relationships, transitional states, change over time, and detailed interactions (McClean et al., 2005). Upon comparing 2D drawings on PowerPoint slides with 3D animations, the 3D lessons were found to be more effective at teaching human anatomy, but only concerning concepts requiring spatial ability (Hoyek et al., 2014).

However, there are many different styles and methods available for animation design. Betrancourt and Tversky (2000) outline two vital principles for animation design: conceptual mapping and concision. In other words, the animation should clearly and simply convey change over time by mapping the information to changes in the display (Betrancourt & Tversky, 2000). One may apply Mayer's signaling principle via a *cuing mechanism* (Lowe & Boucheix, 2011; Mayer, 2008). The signaling may be of an internal nature, such as use of color, or external nature, such as use of bounding boxes or arrows (Lowe & Boucheix, 2011 ; Masakura, Nagai, & Kumada, 2006). However, in complex animations, the use of visuospatial cuing is not very effective, and its efficacy even deteriorates upon multiple viewings (Lowe & Boucheix, 2011).

There is a general assumption that animation is the superior tool for multimedia instruction (Mayer, et al., 2005). However, there are contradictory and inconsistent results over its actual impact on learning: some have found it to be beneficial, some have found it to be detrimental (Betrancourt, 2005; Mayer et al., 2005). A study comparing computer-based animations and paper-based static graphics found that the static graphics group performed better on retention and transfer tests (Mayer et al., 2005). It is not reasonable to ask whether animation is more effective than static graphics as an instructional tool: instead, one must find out *when* and *why* and *for whom* it is so (Betrancourt, 2005). Static graphics and text allow learners to manage intrinsic processing, reduce extraneous processing, and engage germane processing (Mayer et al., 2005). This is because they have control over both pace and order, they see only frames pertinent to each significant stage, and they can explain the transition from one frame to the subsequent one, respectively (Mayer et al., 2005). Animation with narration reduces extraneous processing and engages germane processing (Mayer et al., 2005). It accomplishes this by reducing the working memory required to create mental, verbal, and pictorial representations, removing the effort required to make choices in the learning process and increasing engagement in the learning process (Mayer et al., 2005).

I. <u>The Whiteboard as a Learning Tool</u>

Whiteboard animation is a unique style of 2D animation which has grown in popularity over the past decade, both for the emotional response it invokes and for its novelty. A component of this emotional response is the familiarity with a board in a classroom or clinical environment (Singh et al., 2009). Additionally, whiteboard animation has many innate advantages which may allow it to bridge the gap between static graphics and animations. A common component of whiteboard animation, the presence of a drawing hand has a significant effect on cognition (Castro-Alonso et al., 2014). However, whiteboard animation is not without its weaknesses.

Despite their broad usage and popular reception, whiteboard-style animations have significant limitations. The primary issues with whiteboard animations are: they must often be developed by specialized companies or studios, they are time-consuming, and they can be expensive to produce—although not nearly as expensive as 3D animation (Turkay, 2016). In addition, there appears to be limited experimental evidence showing whether the instructional methods manipulated by whiteboard-style animations increase learning and engagement when compared to other formats (Turkay, 2016). However, it is not necessary for whiteboard animations to be contracted to a specialized studio, with the use of a few easily accessible tools: a camera, a few Expo markers, and a dry erase board.

Dry erase boards are common artefacts in emergency departments, operating theaters, intensive care units, and impatient wards (Wears et al., 2007). They allow for communication both within and between interdependent groups of health professionals (Wears et al., 2007). An emergency department workplace involves quick changes, information-intensive processes, and coordination between a diverse array of healthcare professionals (Bjørn & Hertzum, 2011). Emergency department (ED) employees have found that dry-erase whiteboards are a direct, fast, and flexible way to coordinate, and whiteboards have become central in creating a collaborative environment in high risk, highly dynamic ED spaces (Bjørn & Hertzum, 2011 ; Wears et al., 2007 ; Xiao, Schenkel, Faraj, Mackenzie, & Moss, 2007). In fact, ED clinicians have demonstrated a preference for dry-erase whiteboards over electronic whiteboards (Xiao et al., 2007). The benefits of a whiteboard in a clinical setting are that it is “malleable, ecological, locally owned, widely available, informal, and accessible” (Wears et al., 2007, p. 168 - 169).

The importance of the presence of a board in a learning environment has been iterated by participants in a study comparing Physiology lessons through animation-based learning generated in PowerPoint and PowerPoint lectures: “Board teaching is a must if one must make students understand” (Singh et al., 2009, p. 17). Interactive digital whiteboards have become a prevalent tool in classrooms which attempt to modernize their teaching tools. Interactive whiteboards may increase student attention, save teaching time, and improve student teamwork and discussion. However, interactive whiteboards are expensive and require the cost of training instructors and providing software (Bidaki & Mobasheri, 2013).

2D whiteboard animations are most often videos of a hand drawing images, which may be graphic or symbolic in nature, and writing key statements while a narrator explains a concept. The color scheme is most often simply black and white, although some whiteboard animations feature an accent color for emphasis. For example, Khan Academy has transitioned from a white background to a black background, and uses various bright colors. Whiteboard-style animations can be used to teach scientific and medical lessons, such as the online Khan Academy or Osmosis’ medical student learning platform. They can also be used to share “world-changing ideas” with the general public through resources such as RSA Animate, found at https://www.thersa.org/discover/videos/rsa-animate.

A study done over the course of two years tested knowledge acquisition about infertility using a whiteboard animated video on 400 medical students in their first or second year of training (Thomson et al., 2016). The topic of infertility has many associated myths and has a relatively low knowledge base in the general population (Thomson et al., 2016) making it a topic with a relatively high intrinsic cognitive load (Sweller, 2005). The study using whiteboard visualizations concluded that medical students showed short-term improvements in their knowledge of basic reproductive biology and infertility (Thomson et al., 2016). A whiteboard animation may be a useful instructional tool for communicating lesser-known, complex concepts to health professionals.

There are many common characteristics associated with whiteboard animations which differentiate it from other types of multimedia learning tools. One significant aspect of whiteboard animation is the presence of a human hand drawing the images presented or, in some cases such as Khan Academy, a cursor. The presence of a human hand as a signaling mechanism may direct the learner or may create redundancy in the animation (Sweller, 2005). A study (De Koning & Tabbers, 2013) has shown in 2013 that, in an animation showing lightning formation, an on-screen pointing human hand showing movement resulted in higher retention and transfer performance than when a pointing arrow was used. However, the hand did not actually perform these movements: a static picture of a pointing human hand merely replaced the pointing arrow (De Koning & Tabbers, 2013). Even when compared against the "self-gesture" method, in which participants were shown the animation with the pointing arrow and asked to use their own hand to follow its movement, the on-screen hand group scored higher on retention and transfer tests than the self-gesture group did (De Koning & Tabbers, 2013) . This study was significant because it shows that observing a moving human hand over an animation of non-human dynamic systems can improve learning and cognition of dynamic movement (De Koning & Tabbers, 2013 ; De Koning et al., 2009). This study adds to the existing evidence that observing gestures facilitates learning, aids in comprehension, and helps retention (De Koning & Tabbers, 2013; Marley, Levin, & Glenberg, 2007; Wong et al., 2009).

A study was done to test whether observation of an instructor drawing diagrams significantly facilitates learning (Fiorella & Mayer, 2016). A video-based lesson on the Doppler effect was constructed, with the control group viewing static, completed diagrams as the oral lesson proceeded (Fiorella & Mayer, 2016). In the first experiment, students who listened to the same explanation and were able to observe the instructor's full body as they drew the diagrams scored significantly better on transfer tests than the control group—however, only for students with low prior knowledge (Fiorella & Mayer, 2016). Students with high prior knowledges had no significant difference in scoring (Fiorella & Mayer, 2016). In the second experiment, only the instructor's hand could be seen drawing the diagrams. Students with low prior knowledge or high prior knowledge all scored significantly higher on transfer tests than the control group (Fiorella & Mayer, 2016). In the third experiment, students who watched the diagrams being drawn without the instructor's hand and body did not score higher than the control group (Fiorella & Mayer, 2016). The fourth experiment compared the instructor drawing with their full body in view to drawing with only their hand in view: the group which viewed the hand drawing scored better than the group which viewed the hand and the body, although this improvement was relatively marginal (Fiorella & Mayer, 2016). This study suggests that observing an instructor's hand drawing diagrams promotes the learning process by exploiting the principles of multimedia learning (Fiorella & Mayer, 2016). Furthermore, the drawing hand provides a valuable and motivating social cue (Fiorella & Mayer, 2016).

Another study which positioned various static photos of a grasping hand around abstract symbols showed that recall of the abstract symbols was higher and more accurate for the group which was not provided the images of hands (Castro-Alonso, Ayres, Wong, & Paas, 2018). Still, this can be accounted for by the redundancy and coherence principles of multimedia learning (Mayer, 2008), since the presence of the hand can be classified as unnecessary visual information. Therefore, the presence of a hand has the potential to promote learning, but the principles of multimedia learning and cognitive load theory must be applied for the instructional design to be effective.

Another key feature of whiteboard animation is the style in which it is drawn—often cartoonish, albeit charming, depictions which sacrifice accuracy and detail for the quickness with which they may be drawn, as shown in the online resource 1 Minute Physics (https://www.youtube.com/channel/UCUHW94eEFW7hkUMVaZz4eDg). It has been shown that emotionally appealing graphics may increase knowledge retention (Mayer & Estrella, 2014; Mayer, 2014). However, Mayer has warned about the danger of extraneous processing, which can arise from "seductive" text or superfluous illustrations (Sung & Mayer, 2012). According to the "apprehension principle," for the viewer to learn, the content must be easily and accurately comprehended whereas animations that are too fast, realistic, or detailed will overwhelm the learner (O'Day, 2006 ; Tversky, Morrison, & Betrancourt, 2002)The innate simplicity of whiteboard animation is a great strength in this regard.

Various signaling mechanisms, which are recommended to highlight relevant information (Mayer, 2008), can be employed in the design of whiteboard animations. Zooming in or out, writing out significant words for emphasis, seeing a hand drawing each image, using a single-color accent or color-coding, are all signaling mechanisms in that they focus the learner's attention on the most essential concepts (Richter et al., 2016). Visual signals, as opposed to discursive signals, make verbal and pictorial relationships salient (Richter et al., 2016). However, it has been shown that visuospatial cues in complex animations are not effective and did not produce higher retention scores, with a deteriorating effect on attention direction over subsequent viewings (Lowe & Boucheix, 2011). This may be explained by strength of the attention given to an animation's complex dynamics, which exceeds the strength of effect of visuospatial cues (Lowe & Boucheix, 2011). It was also found that the signaling power of standard color cuing or highlighting the most important entities with a bold red, was superior to anti-cuing, or decreasing the visuospatial importance of less important entities (Lowe & Boucheix, 2011). But perhaps the innate simplicity of a whiteboard animation will prevent the animation's complexity from overpowering internal and external signaling mechanisms.

Another characteristic that renders whiteboard animation endearing is the frequent use of visual metaphors, as shown in learning resources from RSA Animate (https://www.youtube.com/channel/UCvhsiQGy_zcNCiSbeXEjhLg). The metaphor involves presenting new information in terms of a more familiar visual element, thus promoting active learning through the construction of mental models (Carroll & Mack, 1999). This concept of using schema, present in long-term memory to facilitate the integration of new ideas, is present in theories of human cognition (Sweller, 2005), supporting the use of the metaphor in learning has legitimate biological foundations for its success. With the visual metaphor, abstract concepts can be represented through pictorial elements (Tversky et al., 2002). Graphics displays, which can portray both spatial elements and nonspatial elements, possess a clear advantage over written information alone in representing concepts which are not inherently visual or be using metonymy to take advantage of sociocultural visual associations (Tversky et al., 2002). Pictorial languages can even transcend barriers of time and space because of their similarities across cultures (Tversky et al., 2002).

Often, these visual metaphors may be associated with elements of storytelling, as seen in resources by RSA Animate. Storytelling, which is fundamental in clinical environments, is important for learning and can complement the formal learning styles of textbooks and lectures (Calman, 2001). Human communication has historically employed the narrative as a powerful device (Si & Kandel, 2016). Narrative can effectively engage people and facilitate the organization and memorization of information (Si & Kandel, 2016). Narrative structures can emphasize relationships between elements in three discrete ways: it can explain how the subsequent topic is related to the current topic, it can signal a topic transition, and it can create an analogy between the topic and something which exists in the learner's prior knowledge base (Si & Kandel, 2016). Storytelling can even facilitate the interpretation of published scientific work (Phillips, 2012).

However, because whiteboard animation may employ the drawing of static graphics within an animation, these pictorial representations must conform to the Congruence Principle and the Apprehension Principle to be effective (Tversky et al., 2002). According to these principles, the content and format of the graphic should directly correspond to the concepts conveyed, especially change over time, and graphics should be accurate and concise (Tversky et al., 2002). If whiteboard animation is designed to avoid unnecessary complexity or speed and includes interactive elements, it will overcome the disadvantages of animation (Tversky et al., 2002).

Whiteboard animation, if designed purposefully with these principles in mind, can multiply the merits of both static images and animations. Exacerbated by the emotional appeal and familiarity of a whiteboard, a deliberately designed whiteboard-style animation can provide a powerful learning tool.

J. **Research gap**

It has been shown that Mayer's principles of multimedia design improve the learning outcomes of medical students (Issa et al., 2011), and that animation has the potential to increase learning outcomes (Berney & Betrancourt, 2016). However, the impact of whiteboard animation specifically is limited in scope. Whiteboard animation has been frequently used to discern complex concepts by RSA Animate and Khan Academy (https://www.youtube.com/user/khanacademy) to educated audiences. However, best practices for whiteboard animation are loosely and casually defined, if at all, and whiteboard animation design rarely relies on evidence-based research. It has been shown by Cogni+ive (https://www.youtube.com/user/TheCognitiveMedia), the studio responsible for RSA Animate's instructional whiteboard animation videos, that their videos result in greater entertainment and fact retention than merely a video of a talking head. However, it has been long known that "people learn better from words and pictures than words alone" (Mayer, 2005). Ultimately, whether whiteboard animation succeeds in academic or medical education settings is largely unknown and lacks academic research.

III. RESEARCH SIGNIFICANCE

A. Significance of Research Study

This study seeks to analyze the success of 2D whiteboard style animation in comparison to a 3D animation on the topic of long-distance “neuron-like” biofilm communication—a newly elucidated and complex method of chemical communication in bacteria. The benefit of such a study is the examination and comparison of communication styles within the scientific and medical community. Increased engagement and understanding using a whiteboard style animation foster passion in the classroom and potentially lead to increased explorations in research. Not only that, it could garner interest and support within the scientific community as well as the academic community by communicating to various interdependent groups of health professionals, scientists, and clinicians. In addition, whiteboard animation can be much more cost-effective than 3D computer-based animation: it can be done in less time, with fewer resources, and by someone with limited technical expertise. Whiteboard animation has the potential to be another tool scientific and medical animators can add to their professional skillset. The results of this study may show whether whiteboard animation should be taken more seriously as a valid tool for learning success and whether its usage should be expanded within the field of biomedical visualization. Additionally, results from this study may be used to develop and define the best practices for whiteboard animation in biomedical visualization.

B. Research question

Does engagement, visual preference, knowledge transfer, and self-reported cognitive load differ among students in fields related to science and/or medicine when observing a 2D whiteboard animation versus a 3D computer-based animation on the same topic of bacteria biofilm communication?

IV. METHODS

A. Research study design

The intent of this study was to compare a whiteboard 2D animation with a 3D animation and weigh knowledge gained and viewer preference. This information has been gathered via surveys taken both before and after the user views the animation.

In this study, participants 18 years and older have taken an anonymous demographic survey which collected information about their education level, and other demographic information (such as age, gender, etc). The participants also took a spatial reasoning test, Engagement with Animation survey based on the Dallas Museum of Art's Engagement with Art Framework, and a self-reported cognitive load test. Half of the group was randomly assigned to view the 2D animation first, and half was randomly assigned to view the 3D animation first. Before viewing the animation, each group was tasked with a pre-test intended to assess their knowledge about bacterial communication. Each participant could view the animation as many times as they wish. Afterwards, they took the same knowledge assessment test as before in order to assess knowledge gained. Participants also took surveys evaluating how much they enjoyed viewing each animation.

B. Stimulus design plan

Two stimuli depicting electrochemical signaling in biofilms were created to compare different animation styles. The first, a 2D whiteboard style of animation, relies on an accelerated video of a hand drawing simple pictures on a whiteboard surface while a voiceover explains what is being presented. This technique relies on visual metaphor and simplified representations of complex concepts. The second stimulus is a traditional 3D animation which visualizes temporal and spatial relationships as accurately and realistically as possible and creates a more realistic visualization of bacteria and their communication. The purpose of each is to communicate how biofilms communicate via brain-like bursts of electricity.

1. Research and content mapping

Pre-production began with the creation of a content map. After developing the literature review and meeting with Dr. Prindle, the animation's message was distilled into five primary educational directives:

1. Bacterial electrochemical signaling is a novel form of bacterial communication.
2. Biofilm communication is commonly defined by quorum sensing.
3. Bacterial communication is relevant, both clinically and academically.
4. Biofilm formation is a tightly regulated process.
5. Biofilms express a codependent metabolism.

From these educational goals, the most essential elements of information were isolated and organized. The audience's level of prior knowledge was expected to be highly variable, with the majority of the audience having heard of bacterial signaling/communication, but a very small population having a self-reported level of high prior knowledge. The web-based diagramming platform LucidChart was used to organize this information into an easily manipulated hierarchy (as shown in Figure 2). The content map considers the intended audience, the delivery mechanism, the time allotted for each animation, as well as Dr. Prindle's indication of the most integral information.

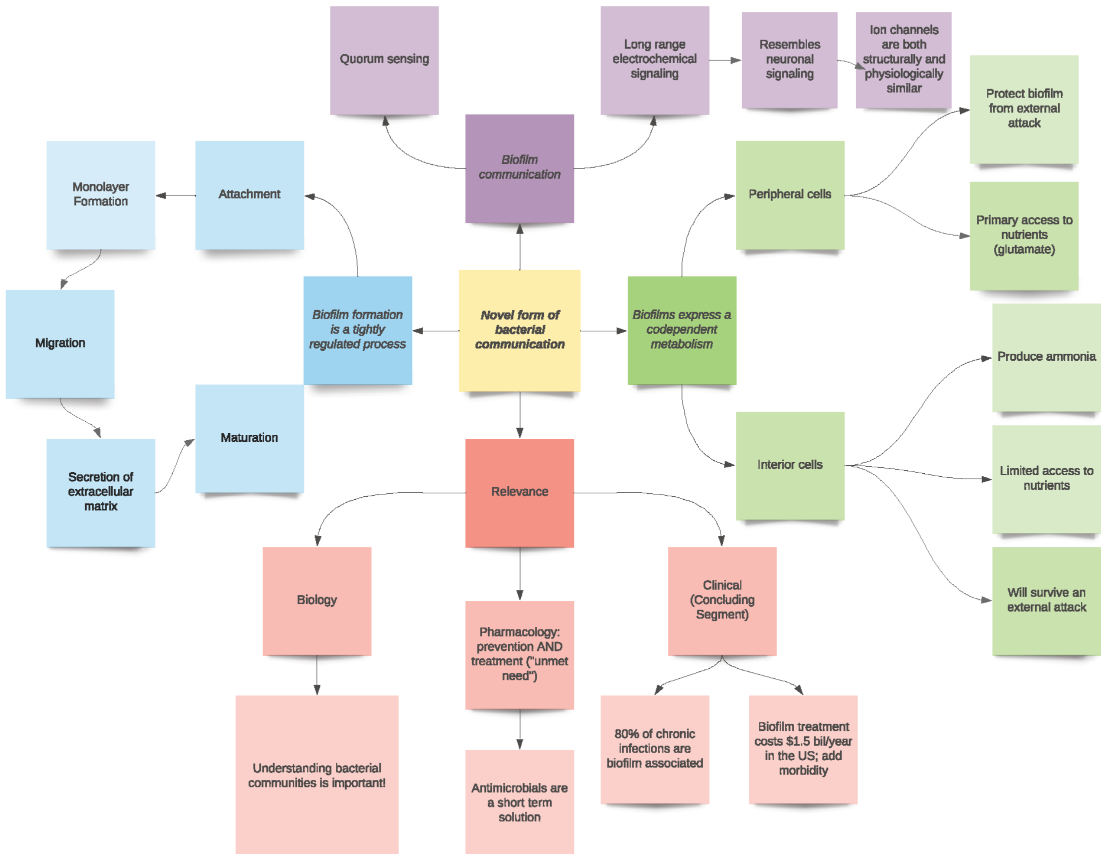


Figure 2. Content map for bacterial electrochemical signaling animations.

The next objective was to align both the 3D animation and whiteboard animation architecture with Mayer's principles of multimedia design (Mayer, 2008) and principles of multimedia for e-learning (Mayer, 2017). Tables 1 and 2, found below, define each principle and describe how each animation would execute that principle.

TABLE I

THE PRINCIPLES OF MULTIMEDIA DESIGN APPLIED TO 3D AND WHITEBOARD ANIMATION

Principle	**Definition**	**Whiteboard Animation**	**3D Animation**
Coherence	Reduce extraneous material.	Only present relevant/key information.	Only present relevant/key information.
Signalling	Highlight essential material.	Key words/values written onscreen.	Key words/values appear onscreen.

Redundancy	Do not add on-screen text to narrated animation.	Do not add on-screen text to narrated animation.	Do not add on-screen text to narrated animation.
Spatial Contiguity	Place printed words next to corresponding graphics.	Place printed words next to corresponding graphics.	Place printed words next to corresponding graphics.
Temporal Contiguity	Present corresponding narration and animation at the same time.	Present corresponding narration and animation at the same time.	Present corresponding narration and animation at the same time.
Segmenting	Present animation in learner-paced segments	Separate drawings segment the story into smaller pieces.	Separate scenes segment the story into smaller pieces.
Pre-training	Provide pretraining in the name, location, and characteristics of key components	Term "electrochemical signaling is introduced before it is explained.	Term "electrochemical signaling is introduced before it is explained.
Modality	Present words as spoken text rather than printed text.	Script is narrated.	Script is narrated.
Multimedia	Present words and pictures rather than words alone.	Accompany animation with narration.	Accompany animation with narration.
Personalization	Present words in conversational style rather than formal style.	Present words in conversational style rather than formal style.	Present words in conversational style rather than formal style.

TABLE II

MULTIMEDIA DESIGN OF E-LEARNING APPLIED TO 3D AND WHITEBOARD ANIMATION

Principle	**Definition**	**Whiteboard Animation**	**3D Animation**
Coherence	Reduce extraneous material.	Only present relevant/key information.	Only present relevant/key information.
Signalling	Highlight essential material.	Key words/values written onscreen.	Key words/values appear onscreen.
Redundancy	Do not add on-screen text to narrated animation.	Do not add on-screen text to narrated animation.	Do not add on-screen text to narrated animation.

Contiguity	Place printed words near any corresponding graphics, and coincide narration with related display	Place printed words near any corresponding graphics, and coincide narration with related display	Place printed words near any corresponding graphics, and coincide narration with related display
Segmenting	Present animation in learner-paced segments	Separate drawings segment the story into smaller pieces.	Separate scenes segment the story into smaller pieces.
Pre-training	Provide pretraining in the name, location, and characteristics of key components	Term "electrochemical signaling is introduced before it is explained.	Term "electrochemical signaling is introduced before it is explained.
Modality	Present words as spoken text rather than printed text.	Script is narrated.	Script is narrated.
Personalization	Present words in conversational style rather than formal style.	Present words in conversational style rather than formal style.	Present words in conversational style rather than formal style.
Voice	Narration should use a human voice rather than a computer voice.	Narration should use a human voice rather than a computer voice.	Narration should use a human voice rather than a computer voice.
Embodiment	Drawing graphics as you explain is more beneficial than explaining a presented drawing as it reflects a real-life social interaction.	Scenes are drawn as they are explained.	Scenes are not drawn as they are explained.

2. Script and storyboard development

Once the content map was finalized, a script and subway storyboard were developed simultaneously. The primary challenge was to begin the animations with an introduction assessing the relevance of this research, and to end the animations with a poignant conclusion reminding the viewer of the severe consequences biofilms have for public health.

The script and storyboard continued to be developed iteratively as Dr. Prindle, my research advisor, and I worked together to develop a term for this novel form of biofilm communication. Ultimately, we arrived on “bacterial electrochemical signaling,” which was an improvement from “bacterial electrophysiology” and “novel neuron-like biofilm communication.” Terms like “bacterial” and “cell to cell” were repeated to emphasize the primary characteristics of bacterial electrochemical signaling.

The storyboard for the 3D animation began as ballpoint pen sketches on drawing paper, which were then toned in grayscale using Adobe Photoshop®, and finally assembled in Adobe InDesign®. The storyboard for the whiteboard animation was sketched onto a 6-up storyboard template, then drawn out in Clip Studio Paint and formatted in Adobe InDesign®.

3. Characteristics of successful whiteboard animation

In order to discern how I would characterize the visual style of my whiteboard animation, I analyzed the most popular whiteboard-style animations. I observed each style’s background color, drawing style, speed, methods, color usage, and narration. I weighed the pros and cons of each characteristic with respect to Mayer’s principles of multimedia learning. Using the Table 3, I rationalized how and why I wanted my whiteboard animation to look.

TABLE III

ANALYSIS OF CHARACTERISTICS OF WHITEBOARD-STYLE ANIMATIONS

CHARACTERISTIC OF A WHITEBOARD-STYLE ANIMATION	RSA ANIMATE (OLD)	RSA ANIMATE (NEW)	KHAN ACADAMY (NEW)	1 MINUTE PHYSICS	PROS/CONS
BACKGROUND COLOR	White	White	Black	White	White is more recognizable. Visibility may be easier if background is white

					for people with limited vision. Black has more of a "modern" look, but modernity/novelty is less important than actual learning impact and accessibility.
DRAWING STYLE	More realistic than symbolic	More realistic than symbolic	Very symbolic. Basic shapes	Very symbolic	Drawings that are more symbolic minimize the information transmitted, reducing cognitive load. However, drawings that are too symbolic do not contain enough information. When it comes to complex topics, drawings must be simple enough to understand, universal enough to be applied to multiple contexts, and complex enough to communicate the concepts being narrated.
(IS IT MORE SYMBOLIC OR MORE REALISTIC?)	Drawing process is sped up in post-production.	Drawing process is much more sped up. Not every illustration is shown to be drawn!	Real time	slightly sped up	If the drawing is sped up too fast, then the point of drawing out each concept is rendered moot, because the drawing is too quick to be managed by the viewer's cognitive capacity. If the drawing is in real time, the drawing may not be completed fast enough to match the narration.
SPEED OF DRAWING (IS IT REAL-TIME OR SPED UP?)	Drawn on actual whiteboard.	Illustration drawn digitally beforehand, and hand is animated on top.	No hand present: digital drawings are shown.	Hand is present. Animation is of marker on paper.	The presence of a drawing hand is *key* for inducing an emotional response in the viewer. Simply viewing a hand drawing can greatly aid in the learning process. Furthermore, I think there is

					something both familiar and comforting about the presence of an actual whiteboard.
ILLUSTRATION METHOD (IS THERE A HAND PRESENT? HOW WAS THE ANIMATION MADE?)	Key words are written out, in all caps.	Key words are written out, but more words are being written out AND speech bubbles are written out as well! (Increase in irrelevant information being written). All words are written in all caps, some in black, some in red.	Some key words are written out. Usually these are labels for whatever is being drawn.	Speech bubbles and sound effects are written out. Sometimes, mathematical formulas are written out (but rarely words)	Writing out key words which correspond to whatever has been/will be drawn can strengthen the integration of auditory and visual information. However, writing too much irrelevant information (or writing out the entire narration) overloads the viewer's cognitive load.
COLOR USAGE	black and red	black and red	multiple bright colors	black, with primary colors as highlight colors	Using a "highlight" color can add depth and complexity. It can act as a signaling mechanism which directs the viewer's attention to keywords and key concepts. However, research shows that for complex animations, signaling mechanisms may have limited and deteriorating effects
ARE WORDS WRITTEN OUT?	Based off of script.	Based off of script.	Narration in conversational format.	Narration in conversational format.	Informal language can aid in the viewer's understanding.

4. Asset creation

Zbrush®, Autodesk 3DS Max®, and Adobe After Effects® were used for asset creation, modeling, and animation of the 3D computer-based animation. A whiteboard, Expo markers, webcam, and Adobe AfterEffects® were used to film and edit the whiteboard style animation. Adobe Audition® was used for the implementation of the audio narration at each animation. The same voiceover script was for each animation to ensure control over as many variables as possible.

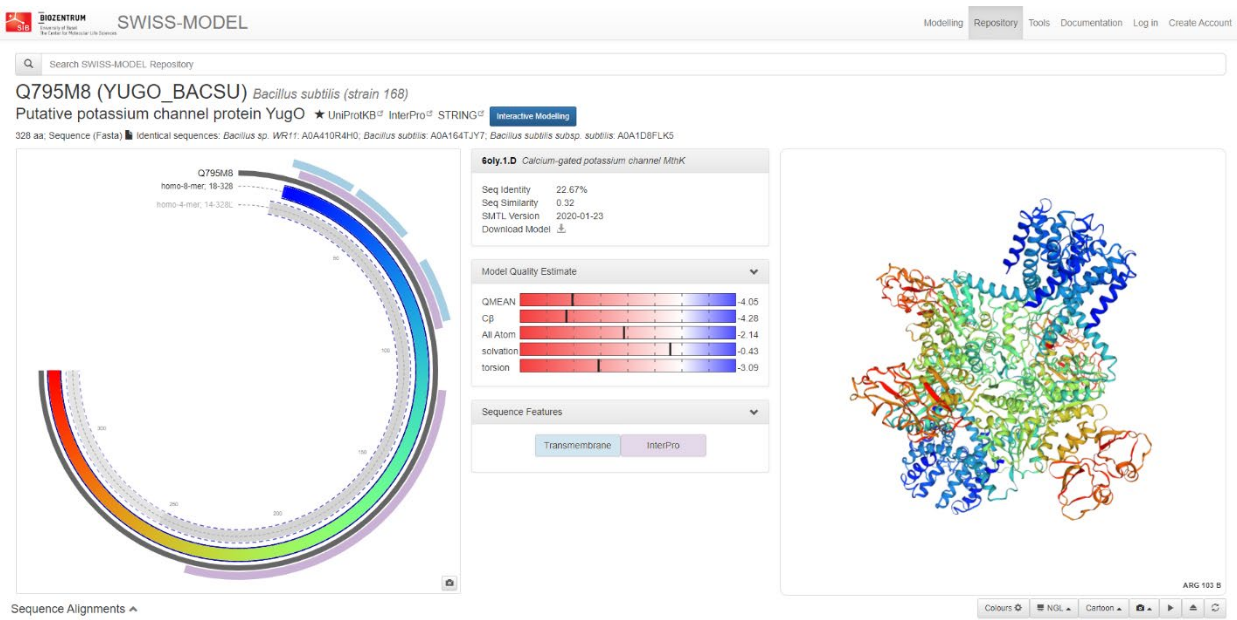


Figure 3. Screenshot of the YugO homology model (Q795M8) obtained from the Swiss-MODEL Repository website.

For the 3D modeling of the YugO potassium ion channel in *Bacillus subtilis,* Swiss-MODEL Repository was accessed to obtain a prebuilt a homology model. This model of the target YugO sequence was built upon a template structure of the homologous MtHK ion channel, another potassium ion channel with significant sequential and structural similarity. Surprisingly, the geometry of the well-studied homologous protein MthK was not merely tetrameric but was that of a tetrameric transmembrane pore with an octameric gating ring of RCK domains (Giraldez & Rothberg, 2017 ; Jiang et al., 2002). Based upon the available literature, we surmised that the geometry of related YugO ion channel could be appropriately represented in a similar way.

Out of eight full-length chains originally available in the homology model, four full-length chains of the homology model containing both the transmembrane pore region and the RCK domains were retained together with four additional partial chains containing only RCK domains. The resulting tetrameric pore structure with octameric gating ring of RCK domains is shown in Figures 4 and 5. VMD was used to create a surface representation of the protein, which was then remeshed in Zbrush®. Figure 4 (below) illustrates the homology model in VMD using a cartoon representation, with each chain a separate color. Figure 5 (below) conveys the surface representation of the YugO homology model, with the full-length chains in blue and the partial RCK domains in white. Using the OPM (Orientation of Proteins in Membranes) Database, the YugO homology model was aligned in a plasma membrane so that its position in the membrane could be visualized for the 3D animation.

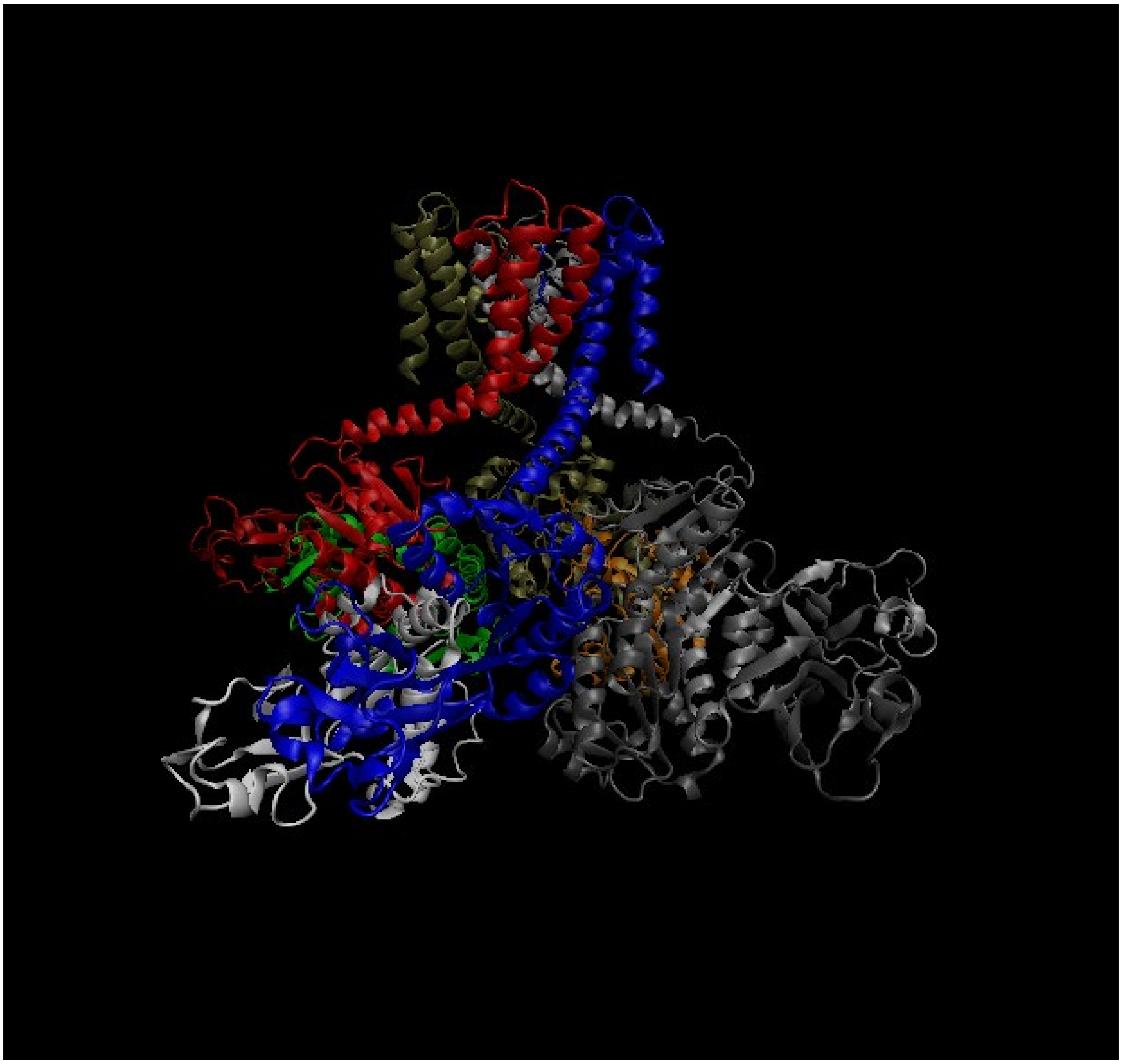


Figure 4. Cartoon representation of YugO in VMD

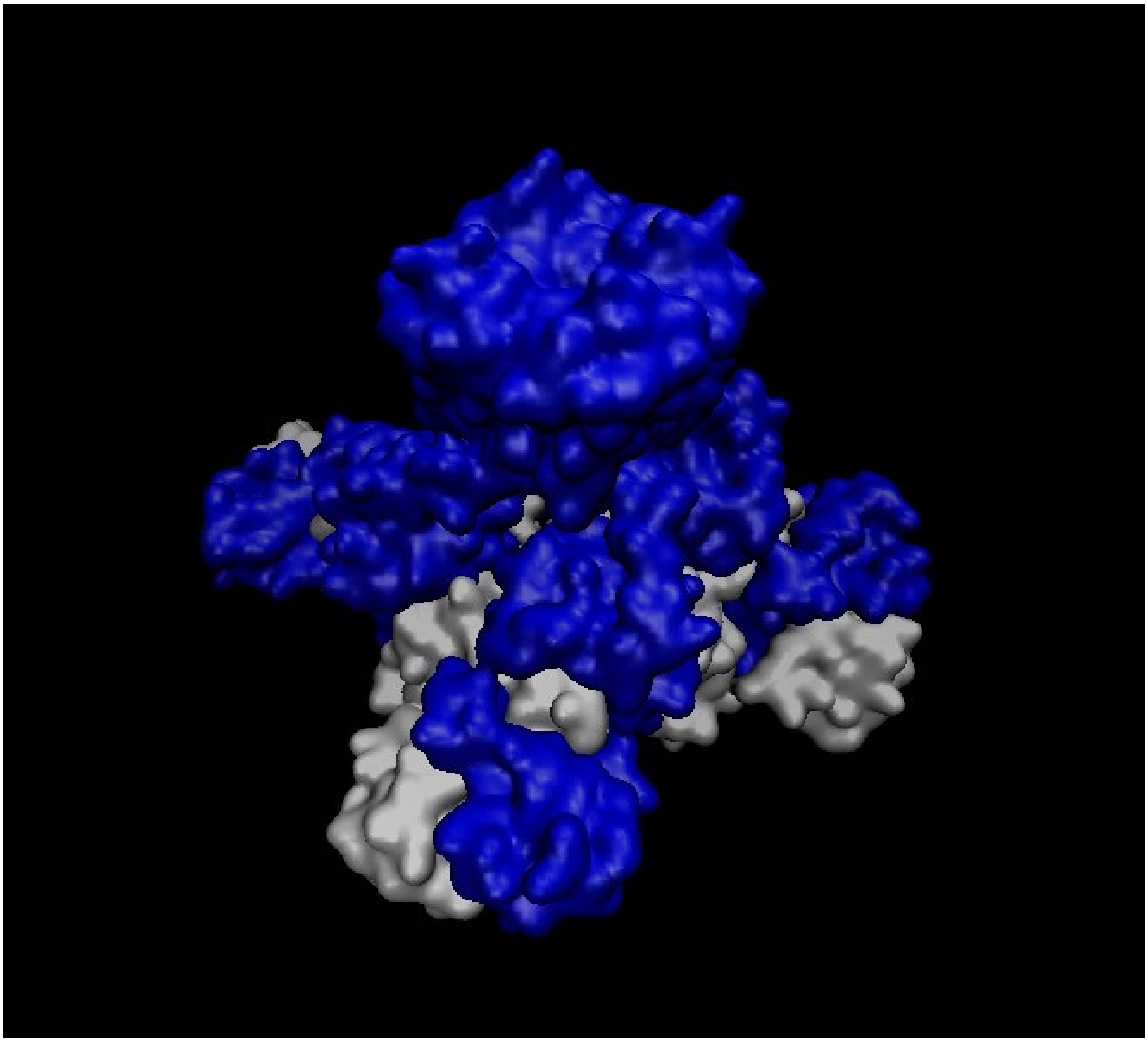


Figure 5. Surf representation of YugO in VMD

The neurons used in the neuron signaling scene were obtained from neuromorpho.org, a site which generates Zbrush® Z spheres in the shape of neurons. The Z spheres were used to generate a mesh, which was Dynameshed and brought into 3DS Max.

5. Animation

The animation of bacterial division and signaling presented some serious challenges, all of which took months of problem-solving, testing, and experimenting. During this time, software updates and changes were accommodated for. Limitations of time, technical expertise, and computing power generated the most impediment.

The first challenge would be to depict bacterial division upon an uneven, organic surface. A plane with a noise modifier on it created an interesting “ground plane” for the bacteria to occupy. Next, splines were aligned to the plane’s surface with an offset of approximately 5 to 10 cm. This offset would allow me to Path Deform capsules to the spline, and animate them moving along it, so that it would appear as if the bacteria were sliding along the surface. Individual division events were generated by animating two capsules on top of each other to move away from each other along the spline, mimicking fission. These capsules were then cloned and keyframed along the spline to give the impression of a chain of bacteria arising from a single cell. These keyframes were then offset from each other in order to generate the appearance of randomness.

The TyFlow plugin for 3DS Max® was used for most of the 3D animation scenes. The Space Colonization growth algorithm was used to “grow” chains of bacteria, which were then bound by a PhysX Joint or PhysX Spring to make the chains rigid. Next, the bacterial chains were subjected to gravity and strewn about the ground plane.

For the first and last scene, which shows a large stationary polymicrobial biofilm, reference objects consisting of cocci (spheres) and capsules (rods) were referenced to generate large, complex bacterial aggregates. TyMesher was then used to extract these meshes to reduce render time.

For the scene exhibiting biofilm growth and electrochemical signaling in the context of a biofilm, a large TyFlow system with 10 different events was created. The first two events were designed to generate a thin blanket of bacteria covering the surface of interest. The eight subsequent events were timed such that every other 30 frames, the biofilm grew. This growth was achieved using TyFlow Spawn. One of the primary challenges with this scene was the computer's inability to perform the massive number of calculations required. As a result, many of the bacteria fell through the assigned ground collider. This problem was fixed by assigning the spawning particles a geometric target: a non-renderable sphere placed directly above the biofilm to attract particles to itself, keeping them from repeatedly colliding with the ground mesh and subsequently falling through. Another solution to this problem was to assign the PhysX Collision of some of the spawning events to Convex Hull rather than Mesh Hull. The Hull of the PhysX Collision identifies the shape of the object against which TyFlow particles collide. Because applying a Mesh Hull can result in miscalculations and objects falling through the collider because of the immense number of processing needed to calculate the event system. A Convex Hull creates a low-poly box-like barrier around the ground collider's reference object. Due to the angle of the camera, it was not immediately obvious that some of the particles were not actually resting on the ground. These solutions allowed for the biofilm to grow iteratively.

During this biofilm growth scene, electrochemical signaling also was animated. The reason for this is because biofilm growth exhibits metabolic oscillations, mediated by electrochemical signaling, as discussed in the literature review. I was unable to implement a render pass of the signaling Light Material over the bacterial surface material via Object ID masks, because I wanted the glow coming off the Light Material to be seen. For me to accomplish this, I had to figure out how to keyframe the material changes within 3DS Max®.

Each of the particles in the 8 spawn events was exported as a TyCache. Next, a VRay Switch Material was keyframed to switch between an orange Light Material and the bacterial surface material. Each of the TyCache events had its own Switch Material, with the keyframes offset so that the signal traveled in and out at the same rate as the oscillating spawn events. This complicated solution was arrived at after many failed attempts to assign the material switches in TyFlow via discrete events. Because TyFlow is a plugin in Beta version which was only just released last year, my own personal technical expertise was severely lacking and led me to attempt creative solutions which could circumvent my lack of knowledge.

6. **Color, materials, and lighting**

In order to develop the look and feel of the 3D animation, I considered the audience and any color conventions in the visual communication of the assets depicted. Because the highlight of the animation was electrochemical signaling, I knew I wanted the steepest value contrast between the bacterial cell’s surface and the signaling. Convention identifies quorum sensing with a fluorescent aqua. Because quorum sensing was used as a point of comparison for electrochemical signaling, I wanted the two hues to be contrasting. By using this rationale, I concluded that dark blue bacterial cells and bright orange electrochemical signaling would communicate the primary message of the animation most effectively. Because the bacterial cells are a dark, saturated blue, the surface upon which they attach and divide would provide the greatest impact to the viewer if it were a warm light gray.

The lighting was arranged in order to complement the hues already selected for the animation’s color palette. To avoid any tinting, the light colors were kept desaturated and of low value. Yellow spotlights and blue fill lights were used in order to create a polished, professional look and feel.

Figure 6. A screenshot of one of the bacterial electrochemical signaling scenes shows the bacterial cells in the biofilm as dark blue, with the electrochemical signaling shown in bright orange.

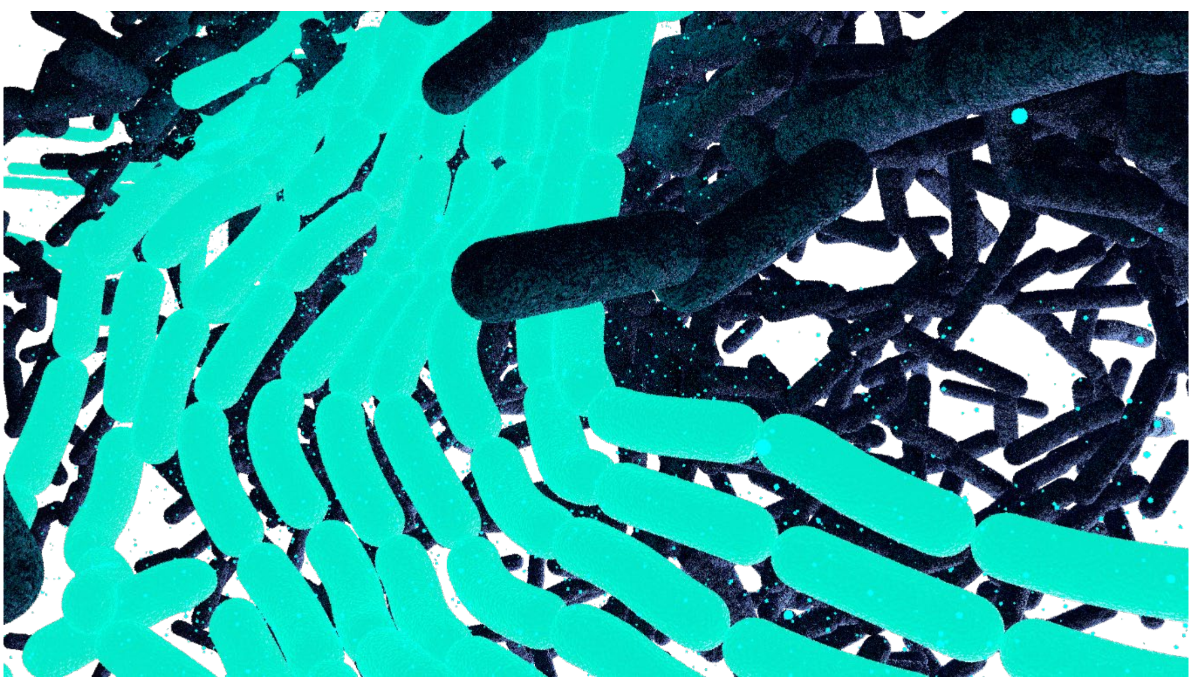


Figure 7. A screenshot of the quorum sensing scene shows both the activated cells and the

extracellular signal as a fluorescent aqua.

The potassium ion channel scene provided a unique challenge, in that it needed to be recognizable as being located on the bacterial surface but could not defocus or detract from any of the other scenes. As a result, the ion channel material was changed from a glass-like, self-illuminated saturated blue to a cool white material which effectively communicated the channel's surface. The bright blue glass material was too distracting, did not communicate the channel's 3D form, and didn't quite fit the established look and feel of the animation. Furthermore, the color of the potassium ions needed to match that of the bacterial electrochemical signaling in order to emphasize the connection between the two scenes and the relationship between these two events.

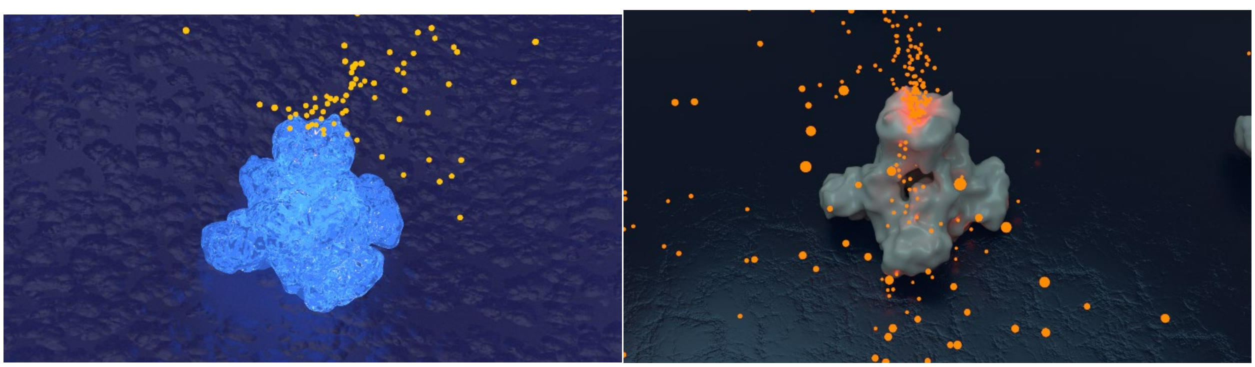


Figure 8. The first, unsuccessful materials and lighting choices for the potassium ion channel (left) and the finalized materials and lighting (right). Note: The 3D structure of the YugO potassium ion channel is not placed correctly. This issue was rectified later in production.

7. Whiteboard animation development

Development of the ideal setup for a whiteboard animation began before the completion of my proposal presentation in July. The first iteration of whiteboard animations consisted of myself drawing on a vertical whiteboard while an assistant took a video recording of the drawing on their phone. Then, these videos were sped up and exported as GIFs via Adobe After

Effects®. The primary issue with this first iteration was the shakiness of the camera, the shakiness of the board, and the challenge of drawing vertically rather than horizontally. Peer critique revealed that the shakiness was a bit distracting, so my next goal was to eliminate any camera shake.

I purchased a 2' by 3' white board, so that I could have more control over the angle I would be drawing at, and a gooseneck desk mount to place my phone on, to avoid camera shake. I recorded myself drawing the faces of my committee members and content advisor using this setup. For isolated drawings, this technique worked perfectly. I then set up professional lights at 45-degree angles to the board in order to properly light the whiteboard while avoiding glare. Placing the whiteboard directly on the floor while using a chair to mount the gooseneck desk mount provided the optimal angle for both drawing and recording with ease. Figures 10 – 12 depict the setup used for the final whiteboard animation.

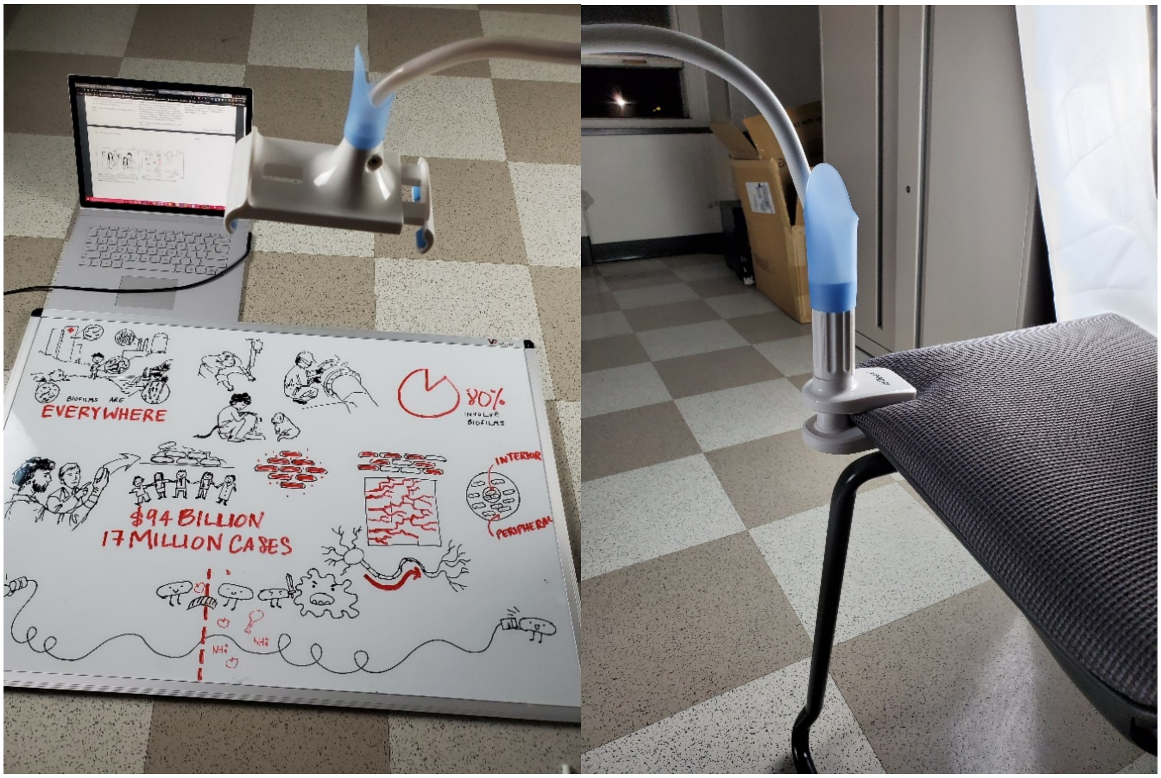


Figure 9. Gooseneck desk mount positioned over whiteboard (left) and securely clamped to chair (right).

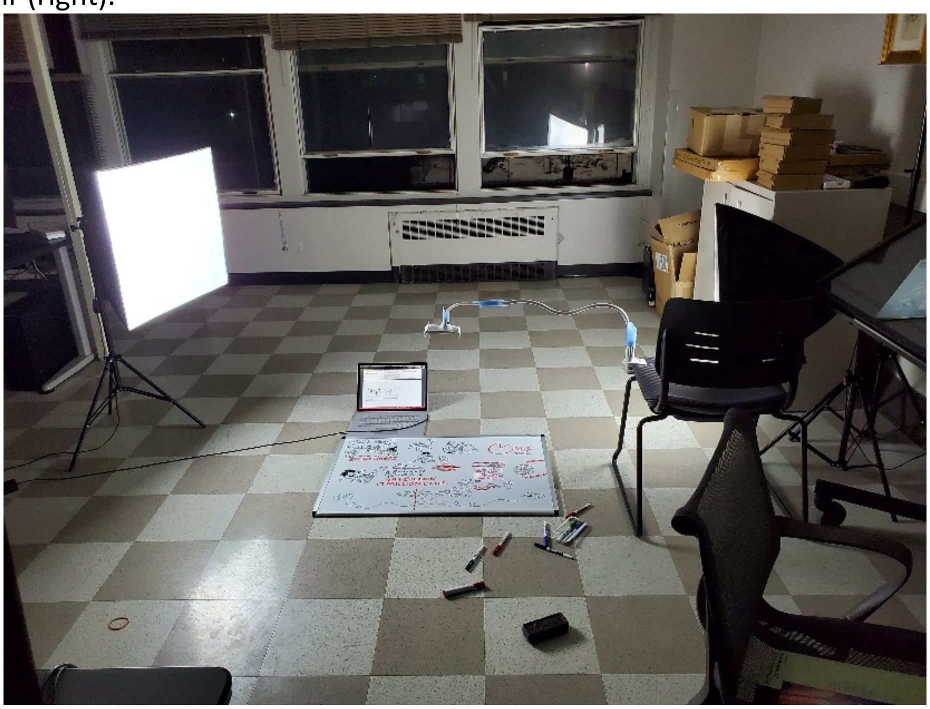


Figure 10. Lighting setup for whiteboard animation.

Figure 11. Workspace setup for whiteboard animation.

Decisions on look and feel were made based on conclusions drawn from the literature review. The drawing hand was shown, and a board with a white background was used. Because it was expected that the audience would have a wide range of prior knowledge concerning bacterial communication and biofilms, only one other color (red) was used in order to reduce cognitive load as much as possible. Too many different colors could distract the viewer from the primary learning objectives. Visual metaphors were also incorporated into the whiteboard animation. Two bacterial cells speaking on tin-can telephones were drawn in a cartoonish style to emphasize that these bacteria have the capability of long-distance communication.

8. IRB Protocol Amendments

Prior to testing, a Claim of Exemption was submitted to the UIC Institutional Review

Board (IRB) and approved on January 22, 2020 under protocol # 2020-0081 (Appendix P). Two amendments, which could have been submitted as one, were later submitted and approved on February 25, 2020. Amendment 1 (Appendix Q) requested to alter the selection strategy. Rather than give a printed handout to students and email them the survey link, it was decided that the link should only be dispersed via email. Amendment 2 (Appendix R) requested to alter the title of the protocol, to adhere to the character limits required for UIC graduate theses. Additionally, the survey comparing both stimuli was modified to reduce bias. The survey both before and after the changes made are shown below for clarity.

Survey Comparing Both Stimuli, Prior to Amendment #2

1. I prefer to use the whiteboard animation of the 3D animation as a learning tool.
    1.1. Strongly disagree
    1.2. Disagree
    1.3. Somewhat disagree
    1.4. Neither agree or disagree
    1.5. Somewhat agree
    1.6. Agree
    1.7. Strongly agree
2. I enjoyed the whiteboard animation more than the 3D animation.
    2.1. Strongly disagree
    2.2. Disagree
    2.3. Somewhat disagree
    2.4. Neither agree or disagree
    2.5. Somewhat agree
    2.6. Agree
    2.7. Strongly agree
3. The whiteboard animation is more successful than the 3D animation at communicating complex information.
    3.1. Strongly disagree
    3.2. Disagree
    3.3. Somewhat disagree
    3.4. Neither agree or disagree
    3.5. Somewhat agree
    3.6. Agree
    3.7. Strongly agree

4. I am more likely to recommend the whiteboard animation than the 3D animation to peers.
    4.1. Extremely unlikely
    4.2. Somewhat likely
    4.3. Unlikely
    4.4. Neither likely nor unlikely
    4.5. Likely
    4.6. Somewhat likely
    4.7. Extremely likely
5. The whiteboard animation was clearer than the 3D animation.
    5.1. Strongly disagree
    5.2. Disagree
    5.3. Somewhat disagree
    5.4. Neither agree or disagree
    5.5. Somewhat agree
    5.6. Agree
    5.7. Strongly agree

Survey Comparing Both Stimuli, Following Amendment #2

I prefer to use the _____ as a learning tool.
- Whiteboard animation
- 3D animation

I enjoyed the _____ most.
- Whiteboard animation
- 3D animation

The _____ is more successful at communicating complex information.
- Whiteboard animation
- 3D animation

I am more likely to recommend the _____ to peers.
- Whiteboard animation
- 3D animation

The ____ was easiest to understand.
- Whiteboard animation
- 3D animation

9. Changes to Qualtrics survey flow

Before the evaluation plan can be discussed, first an error made in the Qualtrics survey flow must be addressed and additional concerns must be acknowledged.

Initially, the survey flow was set so that the knowledge transfer post-test was given to the participant after *both* Stimulus A and B were viewed. The error was rectified by moving the knowledge transfer post-test to be given after the participant viewed the first randomly assigned stimulus. Additionally, the survey questions were not marked as "Force Response" initially. When it was realized that participants could choose not to answer all the survey questions, even if they completed the study and it was marked as "Finished" by UIC Qualtrics, all survey questions were modified to force the participant's response. This modification did not change the wording of the questions of the format of the survey, but it would not allow participants to continue to the next page of the survey without first answering all the questions on that page.

As a result, some of the data may be subject to bias: sections of the survey with the most questions, such as the spatial reasoning test and the knowledge transfer tests, may have been left blank because the user wanted to complete the study as quickly as possible. This may have been the case for some participants acquired through Mechanical Turk, who are participating not because of their own self-interest, but for financial compensation. Further discrepancies may be explained by initial tests run to ensure that the survey flow was as planned, for which Qualtrics would mark as "Finished" but would have no response data.

C. Evaluation plan

This research study will utilize an online delivery using UIC's Qualtrics website through a link provided by the principle investigator and Amazon's Mechanical Turk. Knowledge gained will be tested for this study by comparing pre- and post-test quantitative results. A spatial reasoning test will be provided to assess spatial reasoning ability. Quantitative data will also be gathered using a 7-point Likert scale to understand learner preference, viewer engagement, and self-reported cognitive load. Demographic data acquired via the questionnaires will be utilized for categorizing and comparing populations, noting their level of education, field of study, and familiarity with the topic of bacterial biofilms before seeing the animation. Study participation is voluntary and anonymous.

The data gathered from Qualtrics and Mechanical Turk will be evaluated to compare the learning success and engagement of whiteboard animation among different professional/educational groups: medical students, dental students, pharmacology students, nursing students, graduate science students, and undergraduate students. The purpose of comparing these demographics is to determine which groups are most receptive to a whiteboard-style animated lesson.

1. Study setting

The animation stimulus and pre- and post-test questions will be made available to study participants via a website within the UIC Qualtrics survey framework. Participants may use a computer, laptop, or mobile device to access the study anywhere of their choice. Amazon's Mechanical Turk will be used to obtain additional data from a random sampling laymen and professional population.

2. Sample or population sampling methods

Graduate students at University of Illinois Chicago's College of Medicine and College of Dentistry will be asked to volunteer for the research study. Graduate students of the Biomedical Visualization program will also be asked to participate.

a. Selection criteria

Study participants obtained at UIC are required to be at least 18 years of age and must be students or faculty at UIC in a health professional, biomedical, or science-related field, if asked to participate through the UIC Qualtrics website. The selection criteria for the Amazon Mechanical Turk sample population only need to be over the age of 18, and do not need to be enrolled at UIC.

b. Selection strategy

Participants selected at UIC were given a brief presentation introducing the study during scheduled class time. Initially, it had been planned that printed handouts with the Qualtrics information would be given to students. After IRB submission, it was realized that participants would likely be unwilling to type out a URL and would be more likely to fill out a survey if it was merely emailed to them. As a result, Amendment #1 (Appendix Q) was submitted to the UIC IRB and was approved on February 25, 2020.

After meeting with each of the classes, a link was sent to the class via email with an informed consent form attached. After clicking on the link, participants must read the consent form and give consent to participate in the study. Af A total of 113 responses were recorded in Qualtrics. 90 responses were marked as "Finished" by Qualtrics. Data collection began on February 18, 2020 and was paused on March 14, 2020. Of these 90 completed responses, 63 responses were completed after the survey flow error in Qualtrics was fixed. 35 of these participants were randomly assigned to view the whiteboard animation first. 28 of these participants were randomly assigned to view the 3D animation first. These responses were used to determine if knowledge transfer before and after viewing the animation was statistically significant, if the knowledge transfer between the 3D and the whiteboard animation was statistically significant, and if knowledge transfer had any correlation with spatial reasoning scores, engagement with animation scores, or self-reported cognitive load scores.

The 90 completed responses were analyzed for age, gender, prior education, field of work or study, relevant classes taken, region of residence, viewer preference, and self-reported cognitive load. Qualtrics was used to view a breakout of the data by preference and by self-reported prior knowledge. Qualtrics was also used to generate visualizations comparing viewer responses to the 3D animation and the whiteboard animation.

Although 90 responses were marked as “Finished” by Qualtrics, the number of total responses to each question is not 90. This was explained in Section 8, “Changes to Qualtrics Survey Flow,” and can be attributed to the fact that the survey did not force responses. This is why the demographic data represented in the following figures shows a sample size that is less than 90. ter filling out the consent form, the participants will be directed to a demographic survey, a spatial reasoning test, and knowledge pre-test to establish a baseline of knowledge relevant to bacterial signaling. Any participants who do not fulfill the requirements (at least 18 years old, a student or faculty member in scientific or medical-related fields) or do not complete the study will be removed from the pool. Anyone who does not sign the consent form will not be allowed to participate. The intent of this selection strategy is to obtain correlational data pertaining to viewer engagement, learner preference, and knowledge gained as they may relate to field of study, prior knowledge, spatial reasoning ability, or self-reported cognitive load amongst a health professional or scientific population.

Participants selected via Amazon’s Mechanical Turk will be compensated $3.00 for their time participating in the study. Any participants who do not fulfill the requirements of being over 18 years of age, do not complete the study, or do not fill out the consent form will be removed from the pool. They are not required to be in a scientific or medical related field. After filling out the consent form, the participants will be directed to a demographic survey, a spatial reasoning test, and knowledge pre-test to establish a baseline of knowledge relevant to bacterial signaling. The intent of this selection strategy is to obtain correlational data pertaining to viewer engagement, learner preference, and knowledge gained as they may relate to field of study, prior knowledge, spatial reasoning ability, or self-reported cognitive load amongst a randomly selected population outside of UIC.

After the online survey is completed, participants will have the opportunity to offer feedback and share the survey link with their peers.

c. Size

The anticipated sample size is approximately 500 participants total.

d. Data collection

The data will be collected from completed, anonymous Qualtrics surveys online. Both quantitative data of knowledge gained, spatial reasoning ability, engagement, self-reported cognitive load, and personal preference in combination with qualitative data on demographic information will be collected.

e. Method of analysis

The data will be organized and analyzed in UIC Qualtrics and Microsoft Excel. In UIC Qualtrics, the knowledge transfer data will be obtained by applying the filters “Finished is True” and “Date Recorded after 3/1/2020.” This ensures that only surveys which have been marked as complete are included in data analysis, and the knowledge transfer gathered is *only* from after the Qualtrics survey flow error was fixed. The knowledge transfer data gathered before the discovery and adjustment of the Qualtrics survey flow could not be used because the post test was placed *after* the both stimuli was viewed, not after the first stimulus was viewed.

UIC Qualtrics was also used to make visualizations of the data in the form of tables, pie charts, and bar graphs. UIC Qualtrics also allows the user to breakout a visualization’s data by multiple questions or fields. This feature allows for the analysis of user preference filtered through self-reported Likert-scale prior knowledge levels.

The pre- and post-test scores will be used to calculate knowledge gained. A T-Test for Two Dependent Means will be used to assess if the differences between pre and post-test scores are statistically significant. Knowledge transfer and spatial reasoning tests will be scored, and the percentage of correct answers will be calculated out of 100%. Engagement scores will be translated from the 7-point Likert scale, as shown in Figure 12, to a percentage out of 100%. In a similar manner, the self-reported cognitive load test will be translated from a 5 point scale, as shown in Figure 13, to a percentage out of 100%. These datasets will be analyzed for correlation with each other using Pearson’s Correlation Coefficient will be calculated and analyzed. A T-Test for Two Independent Means will be used to assess whether the difference between knowledge transfer after viewing the whiteboard animation and the knowledge transfer after viewing the 3D animation is statistically significant. Effects size will also be calculated in order to determine how large the differences is.

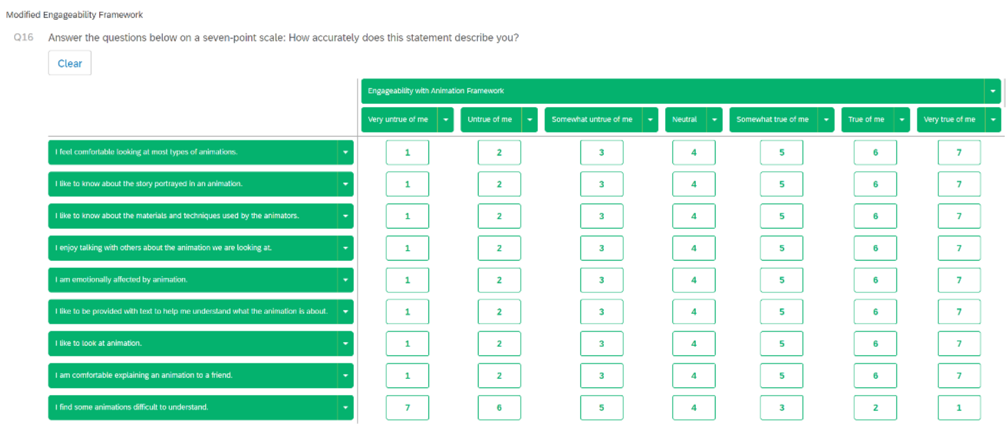


Figure 12. Scoring of Engagement with Animation Framework

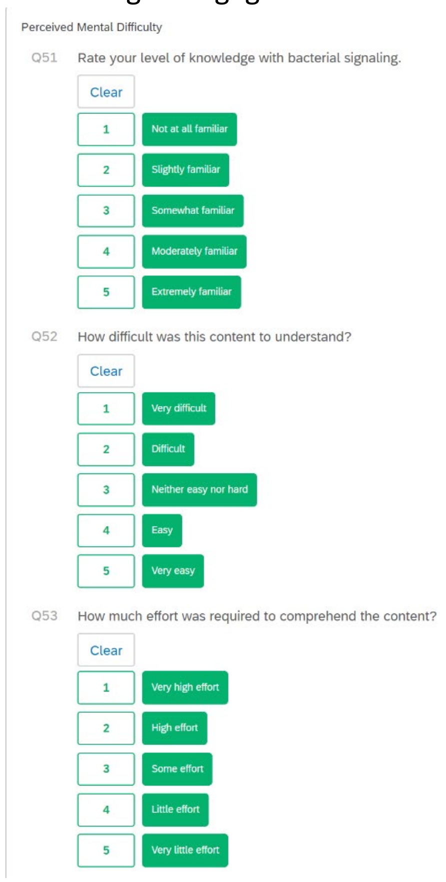


Figure 13. Scoring of Self-Reported Cognitive Load

V. RESULTS

A. Introduction

A total of 113 responses were recorded in Qualtrics. 90 responses were marked as "Finished" by Qualtrics. Data collection began on February 18, 2020 and was paused on March 14, 2020. Of these 90 completed responses, 63 responses were completed after the survey flow error in Qualtrics was fixed. 35 of these participants were randomly assigned to view the whiteboard animation first. 28 of these participants were randomly assigned to view the 3D animation first. These responses were used to determine if knowledge transfer before and after viewing the animation was statistically significant, if the knowledge transfer between the 3D and the whiteboard animation was statistically significant, and if knowledge transfer had any correlation with spatial reasoning scores, engagement with animation scores, or self-reported cognitive load scores.

The 90 completed responses were analyzed for age, gender, prior education, field of work or study, relevant classes taken, region of residence, viewer preference, and self-reported cognitive load. Qualtrics was used to view a breakout of the data by preference and by self-reported prior knowledge. Qualtrics was also used to generate visualizations comparing viewer responses to the 3D animation and the whiteboard animation.

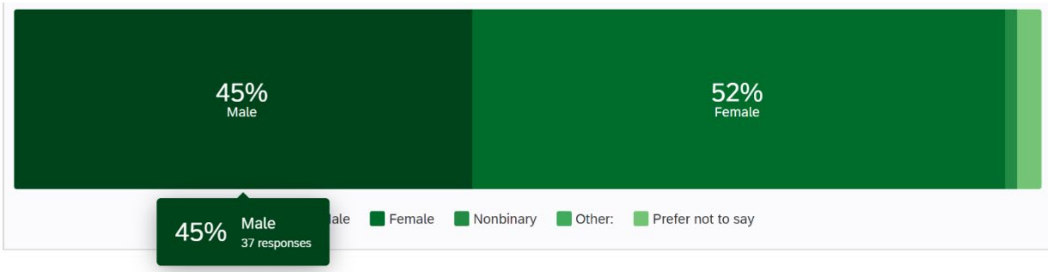
Although 90 responses were marked as "Finished" by Qualtrics, the number of total responses to each question is not 90. This was explained in Section 8, "Changes to Qualtrics Survey Flow," and can be attributed to the fact that the survey did not force responses. Therefore, the demographic data represented in the following section shows a sample size that is less than 90.

B. Expected results

It was expected that there would be a statistically significant amount of knowledge gained after viewing the stimuli, but the knowledge gained would be significantly higher for the whiteboard animation than for the 3D animation. It was expected that there would be a statistically significant correlation between knowledge transfer and spatial reasoning, but there would be a stronger correlation between spatial reasoning and knowledge transfer for participants who viewed the 3D animation first. It was also expected that self-reported cognitive load and engagement would be positively correlated with knowledge transfer.

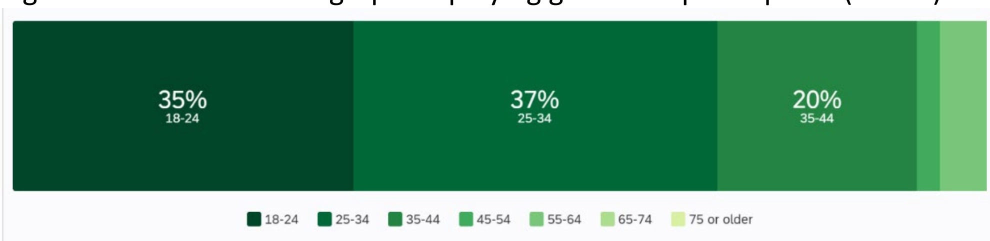
C. Demographic data

The participants surveyed consisted of 37 males, 43 females, and 1 nonbinary participant. Two participants marked “Prefer not to say.” This data is visualized in Figure 14. As shown in Figure 15, 72% of participants were between 18 and 34 years old, with 37% being between ages 25 and 34. Only 6 participants were older than 34. As a result, it did not seem reasonable to conduct any data analysis comparing different age groups.



#	Field		Choice Count
1	Male	44.58%	37
2	Female	51.81%	43
3	Nonbinary	1.20%	1
4	Other:	0.00%	0
5	Prefer not to say	2.41%	2
			83

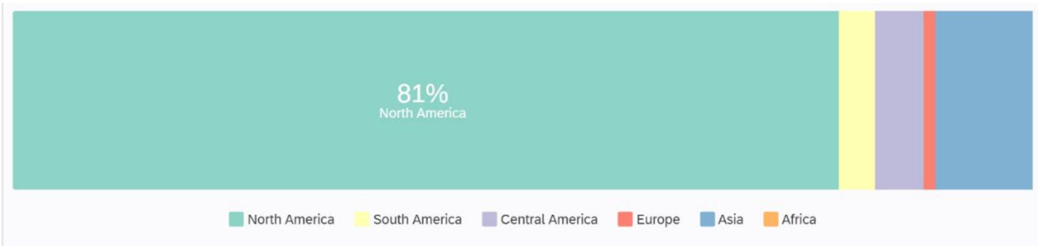
Figure 14. Horizontal bar graph displaying gender of participants (n = 83)



#	Field		Choice Count
7	75 or older	0.00%	0
6	65-74	0.00%	0
5	55-64	4.82%	4
4	45-54	2.41%	2
3	35-44	20.48%	17
2	25-34	37.35%	31
1	18-24	34.94%	29
			83

Figure 15. Horizontal bar graph displaying age groups of participants (n = 83)

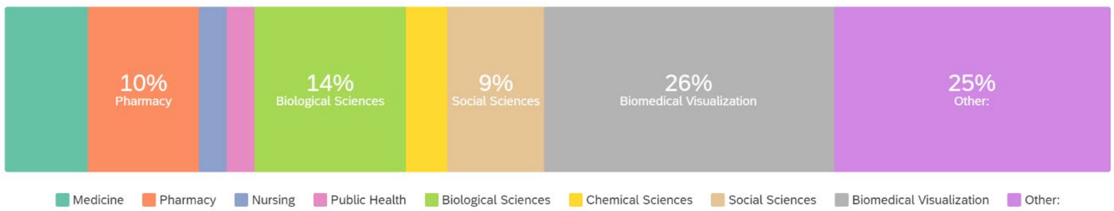
As expected, the majority of participants reside in North America. This data is visualized in Figure 16.



#	Field		Choice Count
1	North America	80.95%	68
2	South America	3.57%	3
3	Central America	4.76%	4
4	Europe	1.19%	1
5	Asia	9.52%	8
6	Africa	0.00%	0
			84

Figure 16. Horizontal bar graph displaying regions of residence of participants (n = 84)

Figure 17, below, displays the largest concentrations of fields of study. Because so many participants were from the program of Biomedical Visualization at UIC (26% of participants), Figure 18, below, excludes BVIS students to show that most participants work or study in the Biological Sciences (14%), Pharmacy (10%), Social Sciences (9%), and Medicine (8%). Most of the fields of work/study marked as "Other" were indicated by the participants to be dentistry or IT. Other less frequent fields of work/study found among participants were agriculture, entertainment, education, government, and environmental science.



#	Field	Choice Count	
1	Medicine	7.50%	6
2	Pharmacy	10.00%	8
3	Nursing	2.50%	2
4	Public Health	2.50%	2
5	Biological Sciences	13.75%	11
6	Chemical Sciences	3.75%	3
7	Social Sciences	8.75%	7
8	Biomedical Visualization	26.25%	21
9	Other:	25.00%	20
			80

Figure 17. Horizontal bar graph displaying field of work/study among participants (n=80)

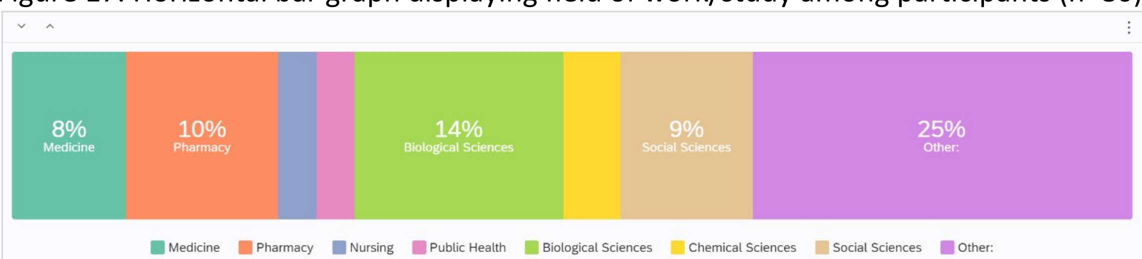
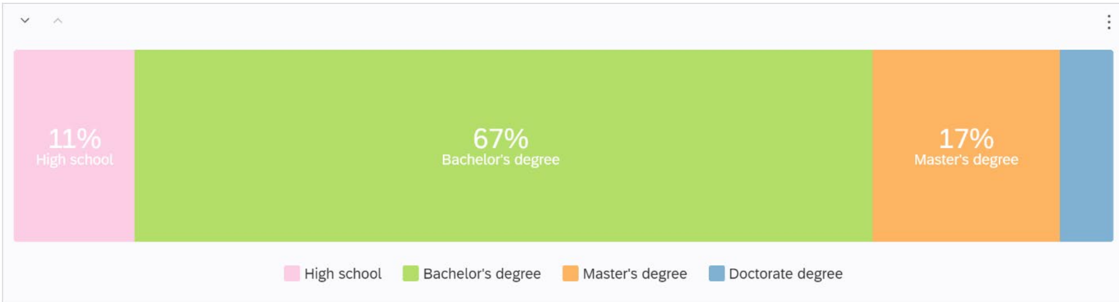


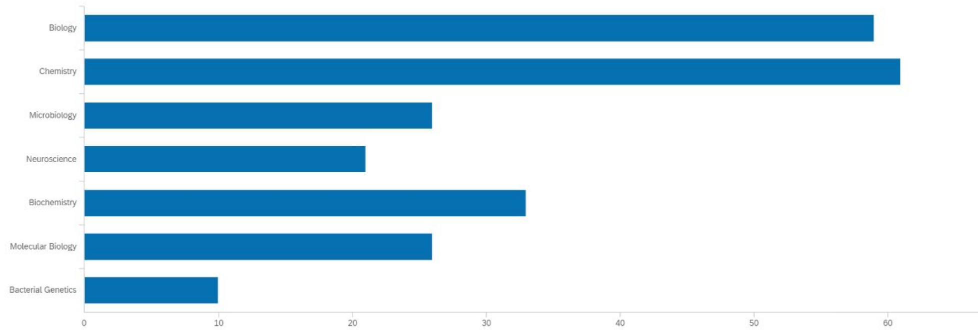
Figure 18. Horizontal bar graph displaying field of work/study among participants, excluding Biomedical Visualization (n=80)

The majority of participants surveyed has a college education, with 67% having a Bachelor's degree. 17% had obtained a Master's and 5% had obtained a PhD. Only 11% had a high school level of education. This indicates that most participants surveyed are well educated and work or study in medical or scientific fields. Additionally, the majority of participants had taken college-level Biology and Chemistry courses. Bacterial Genetics was the least popular course, as only 10 participants had taken it.



#	Field		Choice Count
1	High school	10.98%	9
2	Bachelor's degree	67.07%	55
3	Master's degree	17.07%	14
4	Doctorate degree	4.88%	4
			82

Figure 19. Distribution of education levels of participants (n=82)



#	Field		Choice Count
1	Biology	25%	59
2	Chemistry	26%	61
3	Microbiology	11%	26
4	Neuroscience	9%	21
5	Biochemistry	14%	33
6	Molecular Biology	11%	26
7	Bacterial Genetics	4%	10

Figure 20. Number of participants who have taken college-level courses in these subjects (n=80)

B. Is there a statistically significant increase in knowledge?

In order to move forward with any data analysis comparing 3D animation and whiteboard animation, first it must be determined whether scores improved on the post-test compared to the pre-test, and if this increase in score is statistically significant. In order to do so, a one-tailed T-Test for Two Dependent Means was performed on the pre and post-test scores. The results from the pre-test (M = 39, SD = 17) and post-test (M = 60, SD = 30) knowledge test indicate that, as expected, viewing either of the animations resulted in a statistically significant increase in knowledge, $t(63) = 8.1$, $p < .001$. The calculations are shown below in Table 4.

TABLE IV

ONE-TAILED T-TEST FOR TWO DEPENDENT MEANS ON KNOWLEDGE TRANSFER PRE AND POST-SCORES (n=63)

T-Test for 2 Dependent Means (Pre vs. Post Test Scores)

Treatment 1	**Treatment 2**	**Diff (T2 - T1)**	**Dev (Diff - M)**	**Sq. Dev**
0	0	0	-21	441.05
38.4615385	61.53846154	23.08	2.08	4.31
30.7692308	53.84615385	23.08	2.08	4.31
30.7692308	76.92307692	46.15	25.15	632.65
53.8461538	100	46.15	25.15	632.65
61.5384615	92.30769231	30.77	9.77	95.41
38.4615385	38.46153846	0	-21	441.05
23.0769231	0	-23.08	-44.08	1942.88
30.7692308	15.38461538	-15.38	-36.39	1323.93
23.0769231	15.38461538	-7.69	-28.69	823.32
30.7692308	30.76923077	0	-21	441.05
30.7692308	23.07692308	-7.69	-28.69	823.32
46.1538462	53.84615385	7.69	-13.31	177.13
23.0769231	46.15384615	23.08	2.08	4.31
46.1538462	92.30769231	46.15	25.15	632.65
53.8461538	92.30769231	38.46	17.46	304.86
38.4615385	38.46153846	0	-21	441.05
53.8461538	92.30769231	38.46	17.46	304.86
53.8461538	61.53846154	7.69	-13.31	177.13
38.4615385	46.15384615	7.69	-13.31	177.13
30.7692308	69.23076923	38.46	17.46	304.86
15.3846154	30.76923077	15.38	-5.62	31.55

	46.1538462	61.53846154	15.38	-5.62	31.55
	38.4615385	15.38461538	-23.08	-44.08	1942.88
	46.1538462	61.53846154	15.38	-5.62	31.55
	46.1538462	76.92307692	30.77	9.77	95.41
	69.2307692	76.92307692	7.69	-13.31	177.13
	76.9230769	84.61538462	7.69	-13.31	177.13
	0	0	0	-21	441.05
	61.5384615	76.92307692	15.38	-5.62	31.55
	30.7692308	53.84615385	23.08	2.08	4.31
	61.5384615	100	38.46	17.46	304.86
	46.1538462	92.30769231	46.15	25.15	632.65
	15.3846154	61.53846154	46.15	25.15	632.65
	30.7692308	61.53846154	30.77	9.77	95.41
	38.4615385	76.92307692	38.46	17.46	304.86
	53.8461538	84.61538462	30.77	9.77	95.41
	76.9230769	92.30769231	15.38	-5.62	31.55
	46.1538462	76.92307692	30.77	9.77	95.41
	23.0769231	53.84615385	30.77	9.77	95.41
	15.3846154	53.84615385	38.46	17.46	304.86
	53.8461538	76.92307692	23.08	2.08	4.31
	23.0769231	61.53846154	38.46	17.46	304.86
	23.0769231	69.23076923	46.15	25.15	632.65
	7.69230769	15.38461538	7.69	-13.31	177.13
	30.7692308	30.76923077	0	-21	441.05
	38.4615385	92.30769231	53.85	32.84	1078.79
	23.0769231	69.23076923	46.15	25.15	632.65
	38.4615385	38.46153846	0	-21	441.05
	53.8461538	69.23076923	15.38	-5.62	31.55
	61.5384615	100	38.46	17.46	304.86
	23.0769231	61.53846154	38.46	17.46	304.86
	30.7692308	30.76923077	0	-21	441.05
	46.1538462	69.23076923	23.08	2.08	4.31
	15.3846154	23.07692308	7.69	-13.31	177.13
	61.5384615	53.84615385	-7.69	-28.69	823.32
	30.7692308	69.23076923	38.46	17.46	304.86
	23.0769231	84.61538462	61.54	40.54	1643.27
	30.7692308	92.30769231	61.54	40.54	1643.27
	46.1538462	76.92307692	30.77	9.77	95.41
	46.1538462	30.76923077	-15.38	-36.39	1323.93
	53.8461538	69.23076923	15.38	-5.62	31.55
	53.8461538	76.92307692	23.08	2.08	4.31
Mean	**38.5836386**	**59.58485958**			
Standard Deviation	*17.1166497*	*27.2128417*	M: 21		S: 26533.3

<u>Difference Scores Calculations</u>

Mean: 21
$\mu = 0$
$S2 = SS/df = 26533.3/(63-1) = 427.96$
$S2M = S2/N = 427.96/63 = 6.79$
$SM = \sqrt{S2M} = \sqrt{6.79} = 2.61$

<u>T-value Calculation</u>

$t = (M - \mu)/SM = (21 - 0)/2.61 = 8.06$

$p < 0.00001$

C. <u>Is there a statistically significant difference in knowledge transfer between stimuli?</u>

After determining that participants demonstrated a statistically significant increase in knowledge transfer, the next step was to determine if the knowledge gained after viewing the whiteboard animation was significantly different than the knowledge gained after viewing the 3D animation. This can be determined by performing a one-tailed T-Test of Two Independent Means, and was calculating using socscistatistics.com. In this test, the control group consists of the participants who viewed the 3D animation first. The 35 participants who saw the whiteboard animation first (M = 27, SD = 19) compared to the 28 participants in the control group (M = 14, SD = 20) demonstrated significantly more knowledge gained, $t(63) = 2.5$, $p < .05$. As expected, there is a statistically significant higher participant knowledge transfer after watching the whiteboard animation than participant knowledge transfer after watching the 3D animation. The calculations are shown below in Table 5.

TABLE V
ONE-TAILED T-TEST OF TWO INDEPENDENT MEANS FOR KNOWLEDGE GAINED AFTER WATCHING THE WHITEBOARD ANIMATION AND THE 3D ANIMATION (n=63)

Treatment 1 (X)	Diff (X - M)	Sq. Diff (X - M)^2
0	-26.59	707.21
15.38461538	-11.21	125.64
23.07692308	-3.52	12.37
38.46153846	11.87	140.85
46.15384615	19.56	382.61
46.15384615	19.56	382.61
30.76923077	4.18	17.44
38.46153846	11.87	140.85
30.76923077	4.18	17.44
15.38461538	-11.21	125.64
30.76923077	4.18	17.44
30.76923077	4.18	17.44
38.46153846	11.87	140.85
23.07692308	-3.52	12.37
38.46153846	11.87	140.85
46.15384615	19.56	382.61
7.692307692	-18.9	357.25
0	-26.59	707.21
53.84615385	27.25	742.71
46.15384615	19.56	382.61
0	-26.59	707.21
15.38461538	-11.21	125.64
38.46153846	11.87	140.85
38.46153846	11.87	140.85
0	-26.59	707.21
23.07692308	-3.52	12.37
7.692307692	-18.9	357.25
-7.692307692	-34.29	1175.51
38.46153846	11.87	140.85
61.53846154	34.95	1221.16
61.53846154	34.95	1221.16
30.76923077	4.18	17.44
-15.38461538	-41.98	1762.15
15.38461538	-11.21	125.64
23.07692308	-3.52	12.37
	M: 26.59	SS: 12821.64

Treatment 2 (X)	Diff (X - M)	Sq. Diff (X - M)^2
0	-14.01	-14.01
23.07692308	9.07	9.07
23.07692308	9.07	9.07
46.15384615	32.14	32.14
46.15384615	32.14	32.14
30.76923077	16.76	16.76
0	-14.01	-14.01
-23.07692308	-37.09	-37.09
-15.38461538	-29.4	-29.4

-7.692307692	-21.7	-21.7
0	-14.01	-14.01
-7.692307692	-21.7	-21.7
7.692307692	-6.32	-6.32
23.07692308	9.07	9.07
46.15384615	32.14	32.14
38.46153846	24.45	24.45
0	-14.01	-14.01
38.46153846	24.45	24.45
7.692307692	-6.32	-6.32
7.692307692	-6.32	-6.32
38.46153846	24.45	24.45
15.38461538	1.37	1.37
15.38461538	1.37	1.37
-23.07692308	-37.09	-37.09
15.38461538	1.37	1.37
30.76923077	16.76	16.76
7.692307692	-6.32	-6.32
7.692307692	-6.32	-6.32
23.07692308		
	M: 14.01	M: 14.01
	M: 26.26	SS: 11067.13

<u>Difference Scores Calculations</u>

Treatment 1 (Whiteboard)

N1: 35
df1 = N - 1 = 35 - 1 = 34
M1: 26.59
SS1: 12821.64
s21 = SS1/(N - 1) = 12821.64/(35-1) = 377.11
s1=19.42

Treatment 2 (3D)

N2: 28
df2 = N - 1 = 28 - 1 = 27
M2: 14.01
SS2: 11248.94
s22 = SS2/(N - 1) = 11248.94/(28-1) = 416.63
s2=19.88

T-value Calculation

s2p = ((df1/(df1 + df2)) * s21) + ((df2/(df2 + df2)) * s22) = ((34/61) * 377.11) + ((27/61) * 416.63) = 394.6

s2M1 = s2p/N1 = 394.6/35 = 11.27

s2M2 = s2p/N2 = 394.6/28 = 14.09

t = (M1 - M2)/√(s2M1 + s2M2) = 12.58/√25.37 = 2.5

p = .015193
p < .05
SIGNIFICANT DIFFERENCE BETWEEN CONDITION 1 AND 2

Effect Size Calculator for Independent Samples T-Test (Cohen's d)

Cohen's d = (M2 - M1) ⁄ SDpooled
SDpooled = √((SD12 + SD22) ⁄ 2)

Group 1

Mean (M):
Standard deviation (s):
Sample size (n):

Group 2

Mean (M):
Standard deviation (s):
Sample size (n):

Success!

Cohen's d = (14.01 - 26.59) ⁄ 19.921151 = 0.63149.

Glass's delta = (14.01 - 26.59) ⁄ 19.42 = 0.647786.

Hedges' g = (14.01 - 26.59) ⁄ 19.864284 = 0.633297.
MODERATE POSITIVE DIFFERENCE/EFFECT

There is a statistically significant increase between participant knowledge transfer after watching the 3D animation vs. participant knowledge transfer after watching the whiteboard animation.

D. Is there a correlation between knowledge gained and spatial reasoning?

Using socscistatistics.com, the Pearson's Correlation Coefficient was calculated for knowledge transfer scores out of 100% (x) and spatial reasoning scores out of 100% (y) over a sample size of n=63 participants. Knowledge transfer and spatial reasoning were found to be moderately positively correlated, r(61) = .59, p < .001. There is a significant tendency for high knowledge transfer scores to go with high spatial reasoning scores, as expected.

TABLE VI

PEARSON'S CORRELATION COEFFICIENT FOR KNOWLEDGE TRANSFER SCORES (X) AND SPATIAL REASONING SCORES (Y) (n=63)

X Values	Y Values	X - Mx	Y - My	(X - Mx)2	(Y - My)2	(X - Mx)(Y - My)
0	0	-21.001	-56.236	441.051	3162.468	1181.021
23.07692	85.71429	2.076	29.478	4.309	868.979	61.188
23.07692	71.42857	2.076	15.193	4.309	230.819	31.536
46.15385	85.71429	25.153	29.478	632.655	868.979	741.461
46.15385	85.71429	25.153	29.478	632.655	868.979	741.461
30.76923	57.14286	9.768	0.907	95.414	0.823	8.86
0	28.57143	-21.001	-27.664	441.051	765.319	580.986
-23.0769	57.14286	-44.078	0.907	1942.883	0.823	-39.98
-15.3846	42.85714	-36.386	-13.379	1323.929	178.989	486.795
-7.69231	57.14286	-28.694	0.907	823.319	0.823	-26.026
0	28.57143	-21.001	-27.664	441.051	765.319	580.986
-7.69231	28.57143	-28.694	-27.664	823.319	765.319	793.789
7.692308	28.57143	-13.309	-27.664	177.127	765.319	368.183
23.07692	28.57143	2.076	-27.664	4.309	765.319	-57.423
46.15385	71.42857	25.153	15.193	632.655	230.819	382.137
38.46154	85.71429	17.46	29.478	304.863	868.979	514.703
0	28.57143	-21.001	-27.664	441.051	765.319	580.986
38.46154	57.14286	17.46	0.907	304.863	0.823	15.837
7.692308	28.57143	-13.309	-27.664	177.127	765.319	368.183
7.692308	100	-13.309	43.764	177.127	1915.303	-582.454
38.46154	71.42857	17.46	15.193	304.863	230.819	265.27
15.38462	28.57143	-5.617	-27.664	31.546	765.319	155.38
15.38462	28.57143	-5.617	-27.664	31.546	765.319	155.38
-23.0769	28.57143	-44.078	-27.664	1942.883	765.319	1219.395
15.38462	57.14286	-5.617	0.907	31.546	0.823	-5.094
30.76923	85.71429	9.768	29.478	95.414	868.979	287.946
7.692308	71.42857	-13.309	15.193	177.127	230.819	-202.199
7.692308	28.57143	-13.309	-27.664	177.127	765.319	368.183
0	0	-21.001	-56.236	441.051	3162.468	1181.021
15.38462	71.42857	-5.617	15.193	31.546	230.819	-85.332
23.07692	100	2.076	43.764	4.309	1915.303	90.841
38.46154	85.71429	17.46	29.478	304.863	868.979	514.703
46.15385	85.71429	25.153	29.478	632.655	868.979	741.461
46.15385	71.42857	25.153	15.193	632.655	230.819	382.137
30.76923	85.71429	9.768	29.478	95.414	868.979	287.946
38.46154	100	17.46	43.764	304.863	1915.303	764.136

30.76923	100	9.768	43.764	95.414	1915.303	427.489
15.38462	85.71429	-5.617	29.478	31.546	868.979	-165.569
30.76923	85.71429	9.768	29.478	95.414	868.979	287.946
30.76923	42.85714	9.768	-13.379	95.414	178.989	-130.683
38.46154	71.42857	17.46	15.193	304.863	230.819	265.27
23.07692	71.42857	2.076	15.193	4.309	230.819	31.536
38.46154	28.57143	17.46	-27.664	304.863	765.319	-483.029
46.15385	71.42857	25.153	15.193	632.655	230.819	382.137
7.692308	42.85714	-13.309	-13.379	177.127	178.989	178.056
0	14.28571	-21.001	-41.95	441.051	1759.812	881.004
53.84615	100	32.845	43.764	1078.79	1915.303	1437.431
46.15385	85.71429	25.153	29.478	632.655	868.979	741.461
0	14.28571	-21.001	-41.95	441.051	1759.812	881.004
15.38462	57.14286	-5.617	0.907	31.546	0.823	-5.094
38.46154	28.57143	17.46	-27.664	304.863	765.319	-483.029
38.46154	57.14286	17.46	0.907	304.863	0.823	15.837
0	42.85714	-21.001	-13.379	441.051	178.989	280.969
23.07692	14.28571	2.076	-41.95	4.309	1759.812	-87.076
7.692308	28.57143	-13.309	-27.664	177.127	765.319	368.183
-7.69231	42.85714	-28.694	-13.379	823.319	178.989	383.882
38.46154	57.14286	17.46	0.907	304.863	0.823	15.837
61.53846	100	40.537	43.764	1643.268	1915.303	1774.079
61.53846	57.14286	40.537	0.907	1643.268	0.823	36.768
30.76923	42.85714	9.768	-13.379	95.414	178.989	-130.683
-15.3846	14.28571	-36.386	-41.95	1323.929	1759.812	1526.39
15.38462	57.14286	-5.617	0.907	31.546	0.823	-5.094
23.07692	71.42857	2.076	15.193	4.309	230.819	31.536
		Mx:	My:	Sum:	Sum:	Sum:
		21.001	56.236	26533.296	48519.598	21309.960

Result Details & Calculation

X Values
∑ = 1323.077
Mean = 21.001
∑(X - Mx)2 = SSx = 26533.296

Y Values
∑ = 3542.857
Mean = 56.236
∑(Y - My)2 = SSy = 48519.598

X and Y Combined
N = 63
∑(X - Mx)(Y - My) = 21309.96

R Calculation
r = ∑((X - My)(Y - Mx)) / √((SSx)(SSy))

r = 21309.96 / √((26533.296)(48519.598)) = 0.5939

Meta Numerics (cross-check)
r = 0.5939
MODERATE POSITIVE CORRELATION: tendency for high X variable scores to go with high Y variable scores (and vice versa)
r^2 = .3376527
P Value from Pearson ®
$p < 0.00001$
$p < 0.05$
SIGNIFICANT

E. Is there a correlation between knowledge gained and engagement?

Using socscistatistics.com, the Pearson's Correlation Coefficient was calculated for knowledge transfer scores out of 100% (x) and engagement with animation scores out of 100% (y) over a sample size of n=63 participants. Knowledge transfer and engagement were found to be weakly positively correlated, $r(61) = .25$, $p < .05$. The correlation is not as strong as expected, but it is statistically significant. Table 7 shows the calculations below.

TABLE VII
CALCULATION OF PEARSON'S CORRELATION COEFFICIENT BETWEEN KNOWLEDGE TRANSFER AND ENGAGEMENT (n=63)

X Values	Y Values	X - Mx	Y - My	(X - Mx)2	(Y - My)2	(X - Mx)(Y - My)
0	9.52381	-21.001	-59.839	441.051	3580.676	1256.687
23.07692	85.71429	2.076	16.352	4.309	267.379	33.941

23.07692	58.73016	2.076	-10.632	4.309	113.048	-22.07
46.15385	61.90476	25.153	-7.458	632.655	55.619	-187.583
46.15385	74.60317	25.153	5.241	632.655	27.464	131.815
30.76923	53.96825	9.768	-15.394	95.414	236.985	-150.372
0	85.71429	-21.001	16.352	441.051	267.379	-343.406
-23.0769	69.84127	-44.078	0.479	1942.883	0.229	-21.101
-15.3846	61.90476	-36.386	-7.458	1323.929	55.619	271.358
-7.69231	28.57143	-28.694	-40.791	823.319	1663.916	1170.441
0	74.60317	-21.001	5.241	441.051	27.464	-110.059
-7.69231	61.90476	-28.694	-7.458	823.319	55.619	213.991
7.692308	85.71429	-13.309	16.352	177.127	267.379	-217.624
23.07692	84.12698	2.076	14.764	4.309	217.988	30.647
46.15385	65.07937	25.153	-4.283	632.655	18.346	-107.734
38.46154	74.60317	17.46	5.241	304.863	27.464	91.503
0	73.01587	-21.001	3.653	441.051	13.347	-76.724
38.46154	79.36508	17.46	10.003	304.863	100.05	174.647
7.692308	76.19048	-13.309	6.828	177.127	46.62	-90.872
7.692308	33.33333	-13.309	-36.029	177.127	1298.105	479.51
38.46154	79.36508	17.46	10.003	304.863	100.05	174.647
15.38462	85.71429	-5.617	16.352	31.546	267.379	-91.841
15.38462	71.42857	-5.617	2.066	31.546	4.268	-11.604
-23.0769	47.61905	-44.078	-21.744	1942.883	472.78	958.414

15.38462	65.07937	-5.617	-4.283	31.546	18.346	24.057
30.76923	57.14286	9.768	-12.22	95.414	149.321	-119.362
7.692308	68.25397	-13.309	-1.109	177.127	1.229	14.754
7.692308	85.71429	-13.309	16.352	177.127	267.379	-217.624
0	0	-21.001	-69.363	441.051	4811.165	1456.698
15.38462	87.30159	-5.617	17.939	31.546	321.809	-100.756
23.07692	76.19048	2.076	6.828	4.309	46.62	14.173
38.46154	84.12698	17.46	14.764	304.863	217.988	257.792
46.15385	79.36508	25.153	10.003	632.655	100.05	251.59
46.15385	85.71429	25.153	16.352	632.655	267.379	411.289
30.76923	88.88889	9.768	19.526	95.414	381.278	190.733
38.46154	92.06349	17.46	22.701	304.863	515.332	396.365
30.76923	92.06349	9.768	22.701	95.414	515.332	221.743
15.38462	90.47619	-5.617	21.114	31.546	445.785	-118.587
30.76923	63.49206	9.768	-5.87	95.414	34.463	-57.343
30.76923	71.42857	9.768	2.066	95.414	4.268	20.181
38.46154	76.19048	17.46	6.828	304.863	46.62	119.218
23.07692	100	2.076	30.637	4.309	938.653	63.594
38.46154	76.19048	17.46	6.828	304.863	46.62	119.218
46.15385	63.49206	25.153	-5.87	632.655	34.463	-147.658
7.692308	60.31746	-13.309	-9.045	177.127	81.814	120.38
0	68.25397	-21.001	-1.109	441.051	1.229	23.282

53.84615	65.07937	32.845	-4.283	1078.79	18.346	-140.681
46.15385	73.01587	25.153	3.653	632.655	13.347	91.89
0	50.79365	-21.001	-18.569	441.051	344.804	389.97
15.38462	87.30159	-5.617	17.939	31.546	321.809	-100.756
38.46154	65.07937	17.46	-4.283	304.863	18.346	-74.786
38.46154	71.42857	17.46	2.066	304.863	4.268	36.073
0	79.36508	-21.001	10.003	441.051	100.05	-210.065
23.07692	38.09524	2.076	-31.267	4.309	977.645	-64.902
7.692308	52.38095	-13.309	-16.982	177.127	288.375	226.007
-7.69231	84.12698	-28.694	14.764	823.319	217.988	-423.643
38.46154	34.92063	17.46	-34.442	304.863	1186.246	-601.367
61.53846	61.90476	40.537	-7.458	1643.268	55.619	-302.319
61.53846	96.8254	40.537	27.463	1643.268	754.207	1113.268
30.76923	80.95238	9.768	11.59	95.414	134.324	113.209
-15.3846	80.95238	-36.386	11.59	1323.929	134.324	-421.705
15.38462	50.79365	-5.617	-18.569	31.546	344.804	104.294
23.07692	82.53968	2.076	13.177	4.309	173.637	27.352
		Mx:	My:	Sum:	Sum:	Sum:
		21.001	69.363	26533.296	23520.458	6262.186

Result Details & Calculation
X Values
∑ = 1323.077
Mean = 21.001
∑(X - Mx)2 = SSx = 26533.296

Y Values
∑ = 4369.841
Mean = 69.363
∑(Y - My)2 = SSy = 23520.458

X and Y Combined
N = 63
∑(X - Mx)(Y - My) = 6262.186

R Calculation
r = ∑((X - My)(Y - Mx)) / √((SSx)(SSy))

r = 6262.186 / √((26533.296)(23520.458)) =
0.2507

Meta Numerics (cross-check)
r = 0.2507
WEAK POSITIVE CORRELATION
r^2 = .0629
P Value from Pearson
p=.04572
$p < 0.05$

F. Is there a correlation between knowledge gained and self-reported cognitive load?

Using socscistatistics.com, the Pearson's Correlation Coefficient was calculated for knowledge transfer scores out of 100% (x) and self-reported cognitive load scores out of 100% (y) over a sample size of n=63 participants. Knowledge transfer and engagement were found to be weakly positively correlated, r(61) = .10, p = .42. The correlation is not as strong as expected, not is it statistically significant. Table 8 shows the calculations below.

TABLE VIII
CALCULATION OF PEARSON'S CORRELATION COEFFICIENT BETWEEN KNOWLEDGE TRANSFER AND SELF-REPORTED COGNITIVE LOAD (n=63)

X Values	Y Values	X - Mx	Y - My	(X - Mx)2	(Y - My)2	(X - Mx)(Y - My)

0	0	-21.001	-54.815	441.051	3004.664	1151.178
23.07692	53.33333	2.076	-1.481	4.309	2.195	-3.075
23.07692	66.66667	2.076	11.852	4.309	140.466	24.601
46.15385	66.66667	25.153	11.852	632.655	140.466	298.105
46.15385	60	25.153	5.185	632.655	26.886	130.421
30.76923	60	9.768	5.185	95.414	26.886	50.649
0	53.33333	-21.001	-1.481	441.051	2.195	31.113
-23.0769	53.33333	-44.078	-1.481	1942.883	2.195	65.301
-15.3846	60	-36.386	5.185	1323.929	26.886	-188.667
-7.69231	20	-28.694	-34.815	823.319	1212.071	998.96
0	53.33333	-21.001	-1.481	441.051	2.195	31.113
-7.69231	60	-28.694	5.185	823.319	26.886	-148.781
7.692308	73.33333	-13.309	18.519	177.127	342.936	-246.461
23.07692	73.33333	2.076	18.519	4.309	342.936	38.439
46.15385	46.66667	25.153	-8.148	632.655	66.392	-204.947
38.46154	66.66667	17.46	11.852	304.863	140.466	206.937
0	60	-21.001	5.185	441.051	26.886	-108.895
38.46154	73.33333	17.46	18.519	304.863	342.936	323.339
7.692308	66.66667	-13.309	11.852	177.127	140.466	-157.735
7.692308	33.33333	-13.309	-21.481	177.127	461.454	285.895
38.46154	60	17.46	5.185	304.863	26.886	90.535
15.38462	46.66667	-5.617	-8.148	31.546	66.392	45.765
15.38462	40	-5.617	-14.815	31.546	219.479	83.209
-23.0769	53.33333	-44.078	-1.481	1942.883	2.195	65.301
15.38462	53.33333	-5.617	-1.481	31.546	2.195	8.321
30.76923	60	9.768	5.185	95.414	26.886	50.649
7.692308	73.33333	-13.309	18.519	177.127	342.936	-246.461
7.692308	73.33333	-13.309	18.519	177.127	342.936	-246.461
0	0	-21.001	-54.815	441.051	3004.664	1151.178
15.38462	66.66667	-5.617	11.852	31.546	140.466	-66.567
23.07692	66.66667	2.076	11.852	4.309	140.466	24.601
38.46154	66.66667	17.46	11.852	304.863	140.466	206.937
46.15385	66.66667	25.153	11.852	632.655	140.466	298.105
46.15385	60	25.153	5.185	632.655	26.886	130.421
30.76923	53.33333	9.768	-1.481	95.414	2.195	-14.471
38.46154	66.66667	17.46	11.852	304.863	140.466	206.937
30.76923	73.33333	9.768	18.519	95.414	342.936	180.889
15.38462	60	-5.617	5.185	31.546	26.886	-29.123
30.76923	80	9.768	25.185	95.414	634.294	246.009
30.76923	60	9.768	5.185	95.414	26.886	50.649
38.46154	73.33333	17.46	18.519	304.863	342.936	323.339
23.07692	46.66667	2.076	-8.148	4.309	66.392	-16.913
38.46154	20	17.46	-34.815	304.863	1212.071	-607.878
46.15385	53.33333	25.153	-1.481	632.655	2.195	-37.263
7.692308	40	-13.309	-14.815	177.127	219.479	197.169

0	66.66667	-21.001	11.852	441.051	140.466	-248.903
53.84615	33.33333	32.845	-21.481	1078.79	461.454	-705.558
46.15385	53.33333	25.153	-1.481	632.655	2.195	-37.263
0	60	-21.001	5.185	441.051	26.886	-108.895
15.38462	60	-5.617	5.185	31.546	26.886	-29.123
38.46154	46.66667	17.46	-8.148	304.863	66.392	-142.269
38.46154	33.33333	17.46	-21.481	304.863	461.454	-375.073
0	53.33333	-21.001	-1.481	441.051	2.195	31.113
23.07692	20	2.076	-34.815	4.309	1212.071	-72.265
7.692308	80	-13.309	25.185	177.127	634.294	-335.187
-7.69231	60	-28.694	5.185	823.319	26.886	-148.781
38.46154	53.33333	17.46	-1.481	304.863	2.195	-25.867
61.53846	86.66667	40.537	31.852	1643.268	1014.54	1291.186
61.53846	26.66667	40.537	-28.148	1643.268	792.318	-1141.05
30.76923	60	9.768	5.185	95.414	26.886	50.649
-15.3846	66.66667	-36.386	11.852	1323.929	140.466	-431.24
15.38462	33.33333	-5.617	-21.481	31.546	461.454	120.653
23.07692	46.66667	2.076	-8.148	4.309	66.392	-16.913
		Mx:	My:	Sum:	Sum:	Sum:
		21.001	54.815	26533.296	19683.951	2347.578

Result Details & Calculation

X Values

∑ = 1323.077

Mean = 21.001

∑(X - Mx)2 = SSx = 26533.296

Y Values

∑ = 3453.333

Mean = 54.815

∑(Y - My)2 = SSy = 19683.951

X and Y Combined

N = 63

∑(X - Mx)(Y - My) = 2347.578

R Calculation

r = ∑((X - My)(Y - Mx)) / √((SSx)(SSy))

r = 2347.578 / √((26533.296)(19683.951)) = 0.1027

Meta Numerics (cross-check)

r = 0.1027

WEAK POSITIVE CORRELATION

r^2 = .0105

P Value from Pearson

p=.423159

G. Analysis of those who prefer 3D animation vs. those who prefer whiteboard animation

The next step was to discern whether participants who preferred the whiteboard animation had significantly higher knowledge transfer, engagement, spatial reasoning, and self-reported cognitive load scores than those who preferred the 3D animation. It was expected that participants who preferred the whiteboard animation would have significantly higher knowledge transfer, significantly lower engagement, significantly lower spatial reasoning, and significantly higher self-reported cognitive load scores than participants who preferred the 3D animation. Two-tailed t-tests of independent means were used determine whether the results met these expectations. These calculations are shown in Table 9, 10, 11, and 12, where Treatment 1 indicates participants who preferred the whiteboard animation and Treatment 2 indicates the participants who preferred the 3D animation.

There was no significant effect for knowledge transfer, $t(82) = .88$, $p = .40$, despite those who preferred the whiteboard animation ($M = 27$, $SD = 19$) attaining higher scores than those who preferred the 3D animation ($M = 23$, $SD = 20$). Unexpectedly, whiteboard animation preferers ($M = 66$, $SD = 24$) attained higher spatial reasoning scores than 3D animation preferers ($M = 61$, $SD = 27$), although there was no significant effect, $t(82) = .94$, $p = .35$. As for engagement scores between the two preference groups, no significant effect, $t(82) = -1.12$, $p = .27$, was observed but 3D animation preferers ($M = 76$, $SD = 13$) demonstrated higher scores than whiteboard animation preferers ($M = 72$, $SD = 17$), as predicted. The 40 participants who preferred the 3D animation ($M = 64$, $SD = 17$) compared to the 42 participants who preferred the whiteboard animation ($M = 56$, $SD = 16$) demonstrated significantly higher self-reported cognitive load scores, $t(82) = -2.59$, $p = .01$. This last finding is the most exciting, as it is both statistically significant and matches the predicted result.

TABLE IX

TWO-TAILED T-TEST OF 2 INDEPENDENT MEANS: KNOWLEDGE TRANSFER OF PARTICIPANTS WHO PREFER WHITEBOARD ANIMATION VS THOSE WHO PREFER 3D ANIMATION (n = 82)

Treatment 1 (X)	Diff (X - M)	Sq. Diff (X - M)^2	Treatment 2 (X)	Diff (X - M)	Sq. Diff (X - M)^2
38.46153846	11.54	133.14	38.461538	15.38	236.69
38.46153846	11.54	133.14	46.153846	23.08	532.54
15.38461538	-11.54	133.14	23.076923	0	0
38.46153846	11.54	133.14	38.461538	15.38	236.69
23.07692308	-3.85	14.79	53.846154	30.77	946.75
46.15384615	19.23	369.82	23.076923	0	0
46.15384615	19.23	369.82	53.846154	30.77	946.75
38.46153846	11.54	133.14	38.461538	15.38	236.69
15.38461538	-11.54	133.14	15.384615	-7.69	59.17
38.46153846	11.54	133.14	46.153846	23.08	532.54
46.15384615	19.23	369.82	15.384615	-7.69	59.17
23.07692308	-3.85	14.79	30.769231	7.69	59.17
46.15384615	19.23	369.82	23.076923	0	0
30.76923077	3.85	14.79	38.461538	15.38	236.69
30.76923077	3.85	14.79	15.384615	-7.69	59.17
23.07692308	-3.85	14.79	46.153846	23.08	532.54
30.76923077	3.85	14.79	46.153846	23.08	532.54
30.76923077	3.85	14.79	38.461538	15.38	236.69
23.07692308	-3.85	14.79	38.461538	15.38	236.69
46.15384615	19.23	369.82	30.769231	7.69	59.17
0	-26.92	724.85	0	-23.08	532.54
-23.0769231	-50	2500	0	-23.08	532.54
7.692307692	-19.23	369.82	-7.692308	-30.77	946.75
-15.3846154	-42.31	1789.94	0	-23.08	532.54
53.84615385	26.92	724.85	15.384615	-7.69	59.17
-7.69230769	-34.62	1198.22	23.076923	0	0
46.15384615	19.23	369.82	0	-23.08	532.54
7.692307692	-19.23	369.82	38.461538	15.38	236.69
46.15384615	19.23	369.82	7.6923077	-15.38	236.69
38.46153846	11.54	133.14	-7.692308	-30.77	946.75
38.46153846	11.54	133.14	15.384615	-7.69	59.17
38.46153846	11.54	133.14	61.538462	38.46	1479.29
0	-26.92	724.85	-23.07692	-46.15	2130.18
7.692307692	-19.23	369.82	30.769231	7.69	59.17
23.07692308	-3.85	14.79	-15.38462	-38.46	1479.29
7.692307692	-19.23	369.82	15.384615	-7.69	59.17
38.46153846	11.54	133.14	30.769231	7.69	59.17
38.46153846	11.54	133.14	7.6923077	-15.38	236.69
15.38461538	-11.54	133.14	7.6923077	-15.38	236.69
61.53846154	34.62	1198.22	23.076923	0	0
23.07692308	-3.85	14.79			
15.38461538	-11.54	133.14		M: 23.08	SS: 16094.67

M: 26.92 SS: 14940.83

Difference Scores Calculations

Treatment 1

N1: 42
df1 = N - 1 = 42 - 1 = 41
M1: 26.92
SS1: 14940.83
s21 = SS1/(N - 1) = 14940.83/(42-1) = 364.41

Treatment 2

N2: 40
df2 = N - 1 = 40 - 1 = 39
M2: 23.08
SS2: 16094.67
s22 = SS2/(N - 1) = 16094.67/(40-1) = 412.68

T-value Calculation

s2p = ((df1/(df1 + df2)) * s21) + ((df2/(df2 + df2)) * s22) = ((41/80) * 364.41) + ((39/80) * 412.68) = 387.94

s2M1 = s2p/N1 = 387.94/42 = 9.24
s2M2 = s2p/N2 = 387.94/40 = 9.7

t = (M1 - M2)/√(s2M1 + s2M2) = 3.85/√18.94 = 0.88

TABLE X

TWO-TAILED T-TEST OF 2 INDEPENDENT MEANS: SPATIAL REASONING OF PARTICIPANTS WHO PREFER WHITEBOARD ANIMATION VS THOSE WHO PREFER 3D ANIMATION (n = 82)

Treatment 1 (X)	Diff (X - M)	Sq. Diff (X - M)^2	Treatment 2 (X)	Diff (X - M)	Sq. Diff (X - M)^2
85.71428571	19.73	389.19	71.428571	10.71	114.8
100	34.01	1156.93	100	39.29	1543.37

57.14285714	-8.84	78.21	85.714286	25	625
71.42857143	5.44	29.62	85.714286	25	625
85.71428571	19.73	389.19	85.714286	25	625
57.14285714	-8.84	78.21	71.428571	10.71	114.8
85.71428571	19.73	389.19	71.428571	10.71	114.8
100	34.01	1156.93	85.714286	25	625
71.42857143	5.44	29.62	71.428571	10.71	114.8
85.71428571	19.73	389.19	85.714286	25	625
85.71428571	19.73	389.19	71.428571	10.71	114.8
85.71428571	19.73	389.19	57.142857	-3.57	12.76
71.42857143	5.44	29.62	100	39.29	1543.37
85.71428571	19.73	389.19	100	39.29	1543.37
100	34.01	1156.93	85.714286	25	625
71.42857143	5.44	29.62	85.714286	25	625
85.71428571	19.73	389.19	85.714286	25	625
42.85714286	-23.13	534.96	71.428571	10.71	114.8
71.42857143	5.44	29.62	28.571429	-32.14	1033.16
71.42857143	5.44	29.62	57.142857	-3.57	12.76
28.57142857	-37.41	1399.88	14.285714	-46.43	2155.61
57.14285714	-8.84	78.21	28.571429	-32.14	1033.16
42.85714286	-23.13	534.96	28.571429	-32.14	1033.16
42.85714286	-23.13	534.96	14.285714	-46.43	2155.61
100	34.01	1156.93	57.142857	-3.57	12.76
57.14285714	-8.84	78.21	28.571429	-32.14	1033.16
85.71428571	19.73	389.19	42.857143	-17.86	318.88
28.57142857	-37.41	1399.88	57.142857	-3.57	12.76
71.42857143	5.44	29.62	28.571429	-32.14	1033.16
28.57142857	-37.41	1399.88	42.857143	-17.86	318.88
57.14285714	-8.84	78.21	28.571429	-32.14	1033.16
85.71428571	19.73	389.19	100	39.29	1543.37
28.57142857	-37.41	1399.88	28.571429	-32.14	1033.16
100	34.01	1156.93	42.857143	-17.86	318.88
14.28571429	-51.7	2672.96	14.285714	-46.43	2155.61
28.57142857	-37.41	1399.88	57.142857	-3.57	12.76
57.14285714	-8.84	78.21	85.714286	25	625
71.42857143	5.44	29.62	71.428571	10.71	114.8
28.57142857	-37.41	1399.88	28.571429	-32.14	1033.16
57.14285714	-8.84	78.21	71.428571	10.71	114.8
71.42857143	5.44	29.62			
57.14285714	-8.84	78.21		M: 60.71	SS: 28469.39
	M: 65.99	SS: 23245.87			

Difference Scores Calculations

Treatment 1

N1: 42
df1 = N - 1 = 42 - 1 = 41
M1: 65.99
SS1:
23245.87
s21 = SS1/(N - 1) = 23245.87/(42-1) = 566.97

Treatment 2

N2: 40
df2 = N - 1 = 40 - 1 = 39
M2: 60.71
SS2:
28469.39
s22 = SS2/(N - 1) = 28469.39/(40-1) = 729.98

T-value Calculation

s2p = ((df1/(df1 + df2)) * s21) + ((df2/(df2 + df2)) * s22) = ((41/80) * 566.97) + ((39/80) * 729.98) = 646.44

s2M1 = s2p/N1 = 646.44/42 = 15.39
s2M2 = s2p/N2 = 646.44/40 = 16.16

TABLE XI

TWO-TAILED T-TEST OF 2 INDEPENDENT MEANS: ENGAGEMENT OF PARTICIPANTS WHO PREFER WHITEBOARD ANIMATION VS THOSE WHO PREFER 3D ANIMATION (n = 82)

Treatment 1 (X)	Diff (X - M)	Sq. Diff (X - M)^2	Treatment 2 (X)	Diff (X - M)	Sq. Diff (X - M)^2
61.9047619	-9.9	98.04	90.47619	14.96	223.81
66.66666667	-5.14	26.42	87.301587	11.79	138.9
58.73015873	-13.08	170.99	69.84127	-5.67	32.2
84.12698413	12.32	151.79	84.126984	8.61	74.15
88.88888889	17.08	291.81	69.84127	-5.67	32.2
80.95238095	9.15	83.65	74.603175	-0.91	0.83
80.95238095	9.15	83.65	82.539683	7.02	49.33
85.71428571	13.91	193.43	82.539683	7.02	49.33

87.3015873	15.5	240.1	66.666667	-8.85	78.31
84.12698413	12.32	151.79	92.063492	16.55	273.82
79.36507937	7.56	57.13	76.190476	0.67	0.46
85.71428571	13.91	193.43	92.063492	16.55	273.82
85.71428571	13.91	193.43	76.190476	0.67	0.46
88.88888889	17.08	291.81	92.063492	16.55	273.82
92.06349206	20.26	410.35	90.47619	14.96	223.81
58.73015873	-13.08	170.99	61.904762	-13.61	185.26
63.49206349	-8.31	69.13	74.603175	-0.91	0.83
71.42857143	-0.38	0.14	76.190476	0.67	0.46
100	28.19	794.87	76.190476	0.67	0.46
63.49206349	-8.31	69.13	53.968254	-21.55	464.3
85.71428571	13.91	193.43	68.253968	-7.26	52.74
69.84126984	-1.97	3.86	74.603175	-0.91	0.83
60.31746032	-11.49	132	61.904762	-13.61	185.26
61.9047619	-9.9	98.04	50.793651	-24.72	611.19
65.07936508	-6.73	45.25	87.301587	11.79	138.9
28.57142857	-43.24	1869.27	84.126984	8.61	74.15
73.01587302	1.21	1.46	79.365079	3.85	14.82
85.71428571	13.91	193.43	79.365079	3.85	14.82
65.07936508	-6.73	45.25	76.190476	0.67	0.46
65.07936508	-6.73	45.25	84.126984	8.61	74.15
71.42857143	-0.38	0.14	71.428571	-4.09	16.71
74.6031746	2.8	7.82	61.904762	-13.61	185.26
73.01587302	1.21	1.46	47.619048	-27.9	778.23
33.33333333	-38.47	1480.18	80.952381	5.44	29.56
38.0952381	-33.71	1136.45	80.952381	5.44	29.56
52.38095238	-19.43	377.35	50.793651	-24.72	611.19
34.92063492	-36.89	1360.57	57.142857	-18.37	337.57
79.36507937	7.56	57.13	68.253968	-7.26	52.74
85.71428571	13.91	193.43	85.714286	10.2	104.01
96.82539683	25.02	625.95	100	24.48	599.47
82.53968254	10.73	115.2			
65.07936508	-6.73	45.25		M: 75.52	SS: 6288.17
	M: 71.81	SS: 11770.27			

<u>Difference Scores Calculations</u>

Treatment 1

N1: 42
df1 = N - 1 = 42 - 1 = 41
M1: 71.81

SS1:
11770.27
s21 = SS1/(N - 1) = 11770.27/(42-1) = 287.08

Treatment 2

N2: 40
df2 = N - 1 = 40 - 1 = 39
M2: 75.52
SS2: 6288.17
s22 = SS2/(N - 1) = 6288.17/(40-1) = 161.24

<u>T-value Calculation</u>

s2p = ((df1/(df1 + df2)) * s21) + ((df2/(df2 + df2)) * s22) = ((41/80) * 287.08) + ((39/80) * 161.24) = 225.73

s2M1 = s2p/N1 = 225.73/42 = 5.37
s2M2 = s2p/N2 = 225.73/40 = 5.64

t = (M1 - M2)/√(s2M1 + s2M2) = -3.71/√11.02 = -1.12

TABLE XII

TWO-TAILED T-TEST OF 2 INDEPENDENT MEANS: SELF-REPORTED COGNITIVE LOAD OF PARTICIPANTS WHO PREFER WHITEBOARD ANIMATION VS THOSE WHO PREFER 3D ANIMATION (n = 82)

Treatment 1 (X)	**Diff (X - M)**	**Sq. Diff (X - M)^2**	**Treatment 2 (X)**	**Diff (X - M)**	**Sq. Diff (X - M)^2**
66.66666667	11.11	123.46	46.666667	-18.17	330.03
66.66666667	11.11	123.46	53.333333	-11.5	132.25
53.33333333	-2.22	4.94	100	35.17	1236.69
86.66666667	31.11	967.9	93.333333	28.5	812.25
73.33333333	17.78	316.05	66.666667	1.83	3.36
66.66666667	11.11	123.46	100	35.17	1236.69

73.33333333	17.78	316.05	73.333333	8.5	72.25
40	-15.56	241.98	53.333333	-11.5	132.25
66.66666667	11.11	123.46	100	35.17	1236.69
66.66666667	11.11	123.46	53.333333	-11.5	132.25
66.66666667	11.11	123.46	80	15.17	230.03
53.33333333	-2.22	4.94	60	-4.83	23.36
60	4.44	19.75	66.666667	1.83	3.36
53.33333333	-2.22	4.94	66.666667	1.83	3.36
73.33333333	17.78	316.05	60	-4.83	23.36
66.66666667	11.11	123.46	66.666667	1.83	3.36
80	24.44	597.53	60	-4.83	23.36
60	4.44	19.75	73.333333	8.5	72.25
46.66666667	-8.89	79.01	20	-44.83	2010.03
53.33333333	-2.22	4.94	60	-4.83	23.36
53.33333333	-2.22	4.94	66.666667	1.83	3.36
53.33333333	-2.22	4.94	53.333333	-11.5	132.25
40	-15.56	241.98	60	-4.83	23.36
60	4.44	19.75	60	-4.83	23.36
33.33333333	-22.22	493.83	60	-4.83	23.36
20	-35.56	1264.2	73.333333	8.5	72.25
53.33333333	-2.22	4.94	53.333333	-11.5	132.25
73.33333333	17.78	316.05	73.333333	8.5	72.25
46.66666667	-8.89	79.01	66.666667	1.83	3.36
46.66666667	-8.89	79.01	60	-4.83	23.36
33.33333333	-22.22	493.83	40	-24.83	616.69
66.66666667	11.11	123.46	86.666667	21.83	476.69
60	4.44	19.75	53.333333	-11.5	132.25
33.33333333	-22.22	493.83	60	-4.83	23.36
20	-35.56	1264.2	66.666667	1.83	3.36
80	24.44	597.53	33.333333	-31.5	992.25
53.33333333	-2.22	4.94	60	-4.83	23.36
60	4.44	19.75	73.333333	8.5	72.25
46.66666667	-8.89	79.01	73.333333	8.5	72.25
26.66666667	-28.89	834.57	66.666667	1.83	3.36
46.66666667	-8.89	79.01			
53.33333333	-2.22	4.94		M: 64.83	SS: 10665.56
	M: 55.56	SS: 10281.48			

<u>Difference Scores Calculations</u>

Treatment 1

N1: 42
df1 = N - 1 = 42 - 1 = 41

M1: 55.56
SS1:
10281.48
s21 = SS1/(N - 1) = 10281.48/(42-1) = 250.77

Treatment 2

N2: 40
df2 = N - 1 = 40 - 1 = 39
M2: 64.83
SS2:
10665.56
s22 = SS2/(N - 1) = 10665.56/(40-1) = 273.48

<u>T-value Calculation</u>

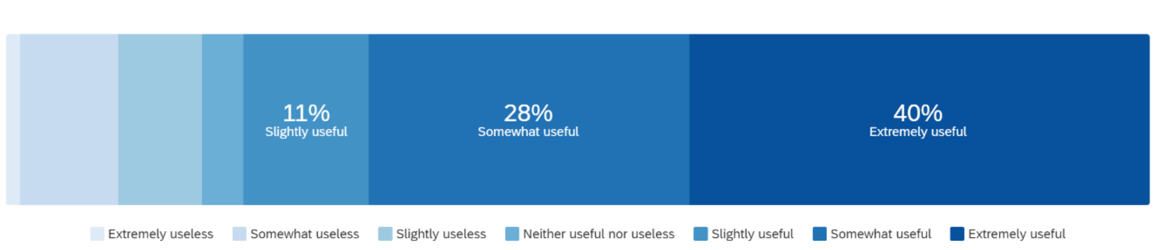
s2p = ((df1/(df1 + df2)) * s21) + ((df2/(df2 + df2)) * s22) = ((41/80) * 250.77) + ((39/80) * 273.48) = 261.84

s2M1 = s2p/N1 = 261.84/42 = 6.23
s2M2 = s2p/N2 = 261.84/40 = 6.55

t = (M1 - M2)/√(s2M1 + s2M2) = -9.28/√12.78 = -2.6

H. Viewer response to 3D animation

Following the viewing of the 3D animation, each participant was asked to answer 5 questions on a 7-point Likert scale. The participant responses were overwhelmingly positive. Most participants found it useful for understanding bacterial signaling. 40% of participants (n=82) found it “Extremely useful” and 28% found it “Somewhat useful.” However, 3% of participants found it “Extremely useless.” 41% of participants strongly agree with the statement “I enjoyed the 3D quality of the animation.” 10% of participants responded that they disagree or somewhat disagree with the statement. 35% strongly agreed with the statement “I enjoyed the complexity of the 3D animation.” When asked, “How likely are you to use 3D animation to communicate your research," 26% of participants responded "Extremely likely" and 21% responded "Neither likely nor unlikely. 40% thought that the information presented in the 3D animation was moderately clear, and 29% thought it was extremely clear. These results are visualized in Figures 21 to 25.

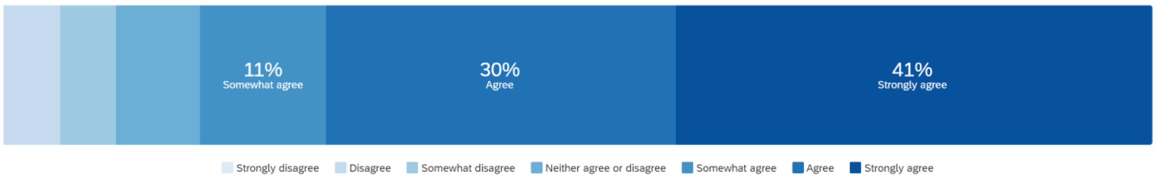


#	Field		Choice Count
2	Somewhat useless	8.54%	7
6	Somewhat useful	28.05%	23
3	Slightly useless	7.32%	6
5	Slightly useful	10.98%	9
4	Neither useful nor useless	3.66%	3
1	Extremely useless	1.22%	1
7	Extremely useful	40.24%	33
			82

Figure 21. 7-point Likert scale response to: How useful is this animation for understanding bacterial electrochemical signaling?

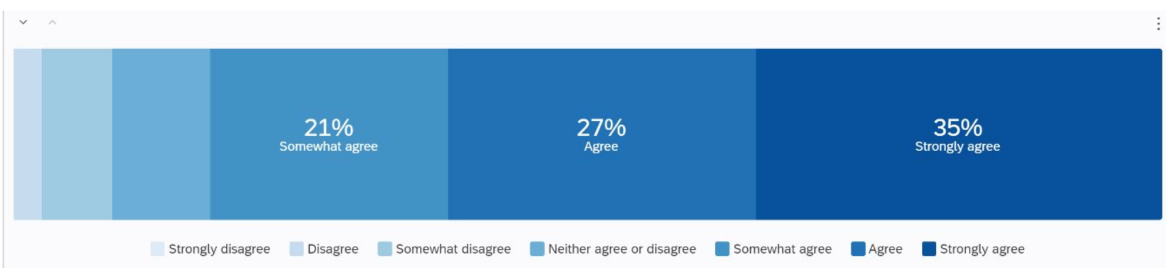
Q42 - How strongly do you agree or disagree with this statement: I enjoyed the 3D quality of the animation.

Page Options



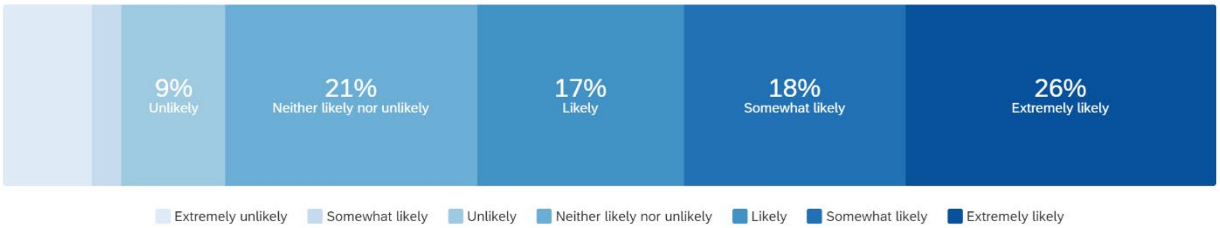
#	Field	Choice Count	
1	Strongly disagree	0.00%	0
7	Strongly agree	41.46%	34
3	Somewhat disagree	4.88%	4
5	Somewhat agree	10.98%	9
4	Neither agree or disagree	7.32%	6
2	Disagree	4.88%	4
6	Agree	30.49%	25
			82

Figure 22. 7-point Likert response to: How strongly do you agree or disagree with this statement: I enjoyed the 3D quality of the animation.



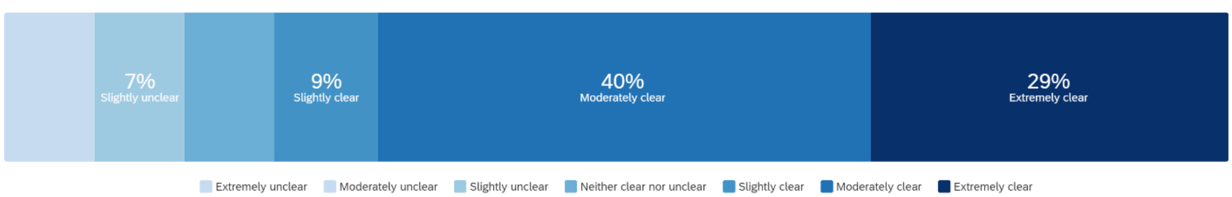
#	Field	Choice Count	
1	Strongly disagree	0.00%	0
2	Disagree	2.44%	2
3	Somewhat disagree	6.10%	5
4	Neither agree or disagree	8.54%	7
5	Somewhat agree	20.73%	17
6	Agree	26.83%	22
7	Strongly agree	35.37%	29
			82

Figure 23. 7-point Likert response to: How strongly do you agree or disagree with this statement: I enjoyed the complexity of the 3D animation.



#	Field	Choice Count	
3	Unlikely	8.54%	7
6	Somewhat likely	18.29%	15
2	Somewhat likely	2.44%	2
4	Neither likely nor unlikely	20.73%	17
5	Likely	17.07%	14
1	Extremely unlikely	7.32%	6
7	Extremely likely	25.61%	21
			82

Figure 24. 7-point Likert response to: How likely are you to use 3D animation to communicate your research?

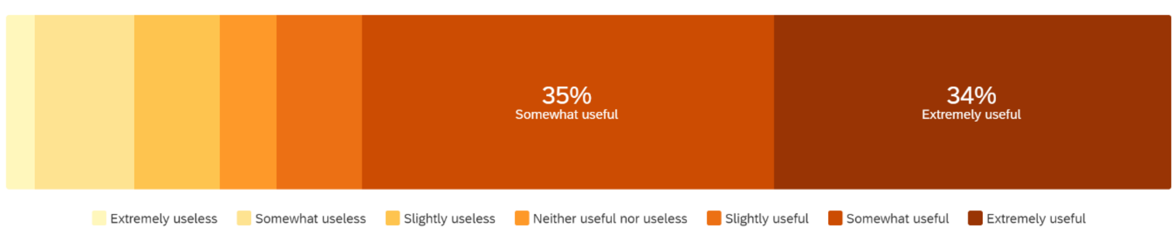


#	Field	Choice Count	
1	Extremely clear	29.27%	24
2	Moderately clear	40.24%	33
3	Slightly clear	8.54%	7
4	Neither clear nor unclear	7.32%	6
5	Slightly unclear	7.32%	6
6	Moderately unclear	4.88%	4
7	Extremely unclear	2.44%	2
			82

Figure 25. 7-point Likert response to: How clear was the information presented in this animation?

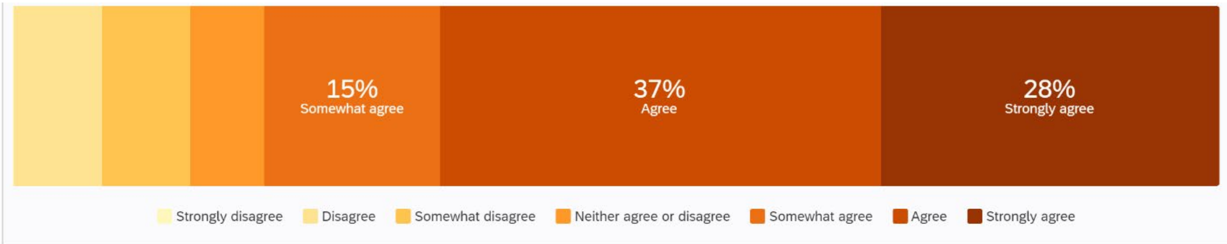
I. Viewer response to whiteboard animation

Following the viewing of the whiteboard animation, each participant was asked to answer 5 questions on a 7-point Likert scale. The participant responses were overwhelmingly positive, although not as positive as the responses to the 3D animation. Most participants found it useful for understanding bacterial signaling: 34% found it extremely useful and 35% found it somewhat useful. 28% of participants strongly agree with the statement "I enjoyed the 2D quality of the animation." 10% of participants responded that they disagree or somewhat disagree with the statement. 32% strongly agreed with the statement "I enjoyed the simplicity of the 2D animation," but 8% strongly disagreed. When asked, "How likely are you to use 3D animation to communicate your research," 23% of participants responded "Somewhat likely" and 20% responded "Neither likely nor unlikely. 35% thought that the information presented in the 3D animation was moderately clear, and 29% thought it was extremely clear. These results are visualized in Figures 26 to 30.



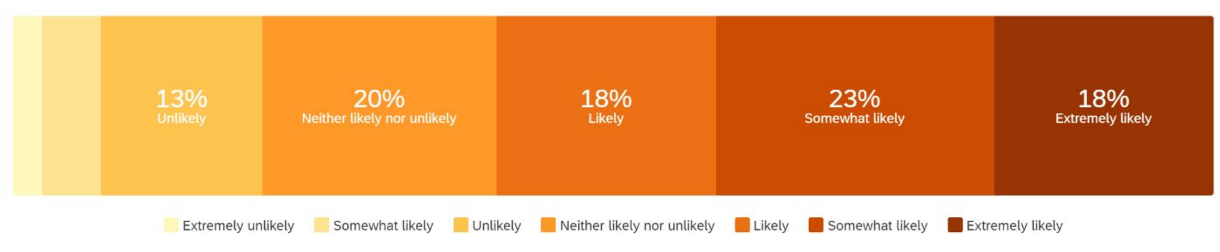
#	Field	Choice Count	
1	Extremely useless	2.44%	2
2	Somewhat useless	8.54%	7
3	Slightly useless	7.32%	6
4	Neither useful nor useless	4.88%	4
5	Slightly useful	7.32%	6
6	Somewhat useful	35.37%	29
7	Extremely useful	34.15%	28
			82

Figure 26. 7-point Likert scale response to: How useful is this animation for understanding bacterial electrochemical signaling?



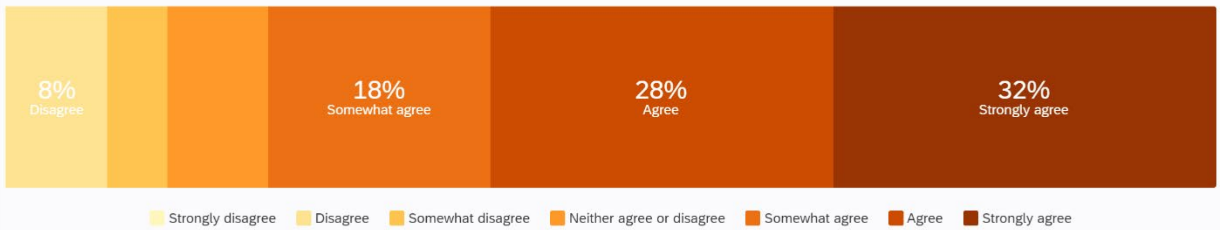
#	Field		Choice Count
1	Strongly disagree	0.00%	0
7	Strongly agree	28.05%	23
3	Somewhat disagree	7.32%	6
5	Somewhat agree	14.63%	12
4	Neither agree or disagree	6.10%	5
2	Disagree	7.32%	6
6	Agree	36.59%	30
			82

Figure 27. 7-point Likert scale response to: How strongly do you agree or disagree with this statement: I enjoyed the 2D quality of the whiteboard animation.



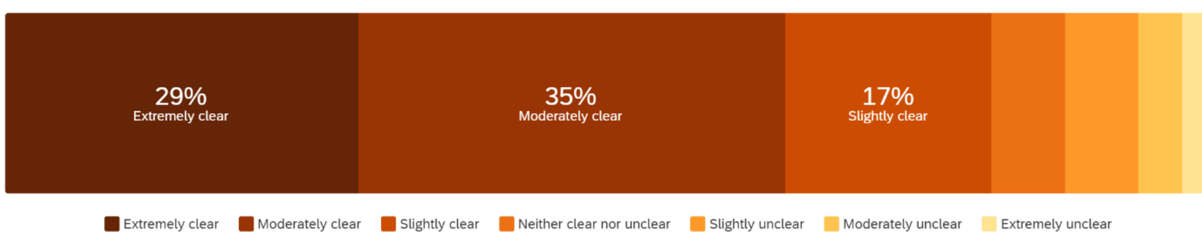
#	Field		Choice Count
1	Extremely unlikely	2.44%	2
2	Somewhat likely	4.88%	4
3	Unlikely	13.41%	11
4	Neither likely nor unlikely	19.51%	16
5	Likely	18.29%	15
6	Somewhat likely	23.17%	19
7	Extremely likely	18.29%	15
			82

Figure 28. 7-point Likert scale response to: How likely are you to use whiteboard animation to communicate your research?



#	Field	Choice Count	
1	Strongly disagree	0.00%	0
2	Disagree	8.33%	5
3	Somewhat disagree	5.00%	3
4	Neither agree or disagree	8.33%	5
5	Somewhat agree	18.33%	11
6	Agree	28.33%	17
7	Strongly agree	31.67%	19
			60

Figure 29. 7-point Likert response to: How strongly do you agree or disagree with this statement: I enjoyed the simplicity of the 2D animation.



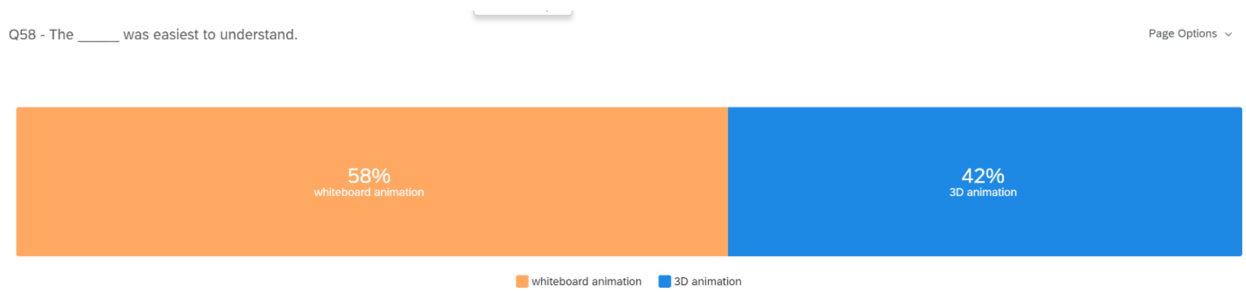
#	Field	Choice Count	
5	Slightly unclear	6.10%	5
3	Slightly clear	17.07%	14
4	Neither clear nor unclear	6.10%	5
6	Moderately unclear	3.66%	3
2	Moderately clear	35.37%	29
7	Extremely unclear	2.44%	2
1	Extremely clear	29.27%	24
			82

Figure 30. 7-point Likert response to: How clear was the information presented in this

animation?

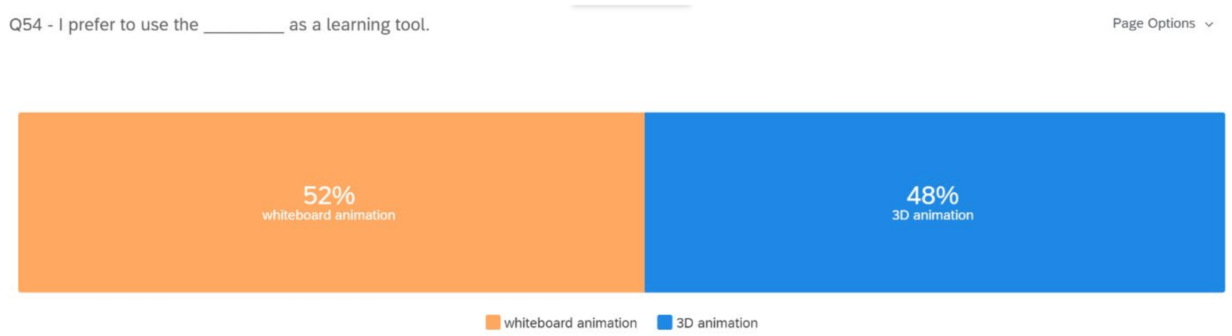
J. Viewer preference between 3D and whiteboard animation

Figures 31 through 35 indicate which animation participants preferred more: whiteboard or 3D animation? What is most intriguing is that although 58% of participants responded that the whiteboard animation was easiest to understand, the preference for learning tool was nearly evenly divided between whiteboard (52%) and 3D animation (48%). Even more interesting, 60% of participants enjoyed the 3D animation more, 53% indicating that the 3D animation is more successful at communicating complex information, and 57% are more likely to recommend the 3D animation to peers.



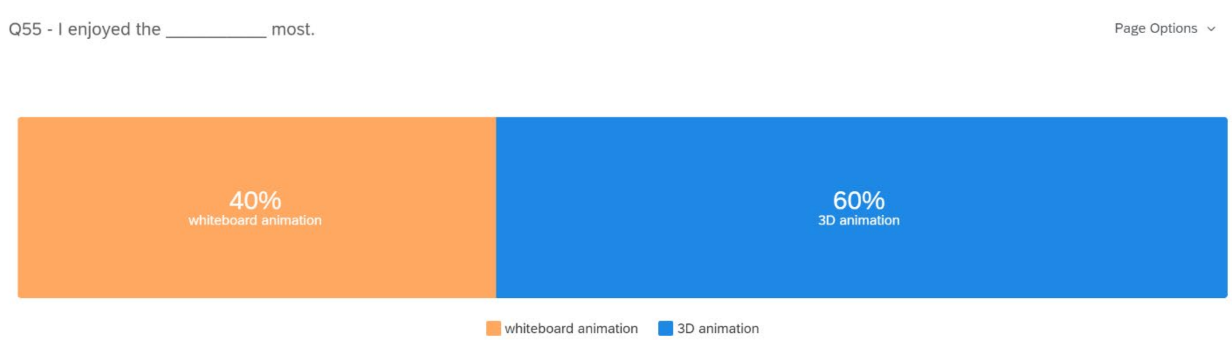
#	Field	Choice Count	
1	whiteboard animation	58.02%	47
2	3D animation	41.98%	34
			81

Figure 31. Participant response to Q: “The _____ was easiest to understand.”



#	Field		Choice Count
1	whiteboard animation	51.85%	42
2	3D animation	48.15%	39
			81

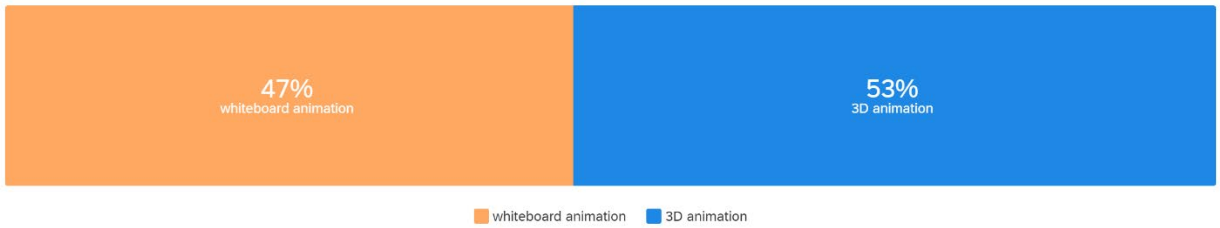
Figure 32. Participant response to Q: “I prefer to use the ____ as a learning tool.”



#	Field		Choice Count
1	whiteboard animation	39.51%	32
2	3D animation	60.49%	49
			81

Figure 33. Participant response to Q: “I enjoyed the ____ most.”

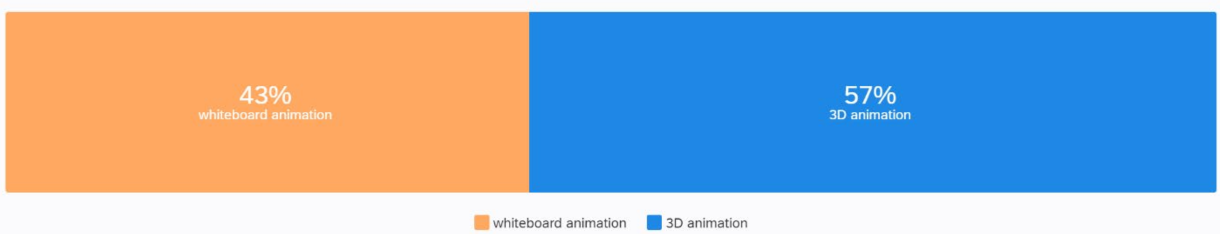
Q56 - The _________ is more successful at communicating complex information.



#	Field	Choice Count	
1	whiteboard animation	46.91%	38
2	3D animation	53.09%	43
			81

Figure 34. Participant response to Q: "The _____ is more successful at communicating complex information."

Q57 - I am more likely to recommend the __________ to peers.



#	Field	Choice Count	
1	whiteboard animation	43.21%	35
2	3D animation	56.79%	46
			81

Figure 35. Participant response to Q: "I am more likely to recommend the ___ to peers."

DISCUSSION

K. **Breakdown by viewer preference**

In order to determine the characteristics of participants who preferred the whiteboard animation and those who preferred the 3D animation as a learning tool, UIC Qualtrics was used to apply a report breakout by these preferences. Figures 36 – 47 show the information gathered from this breakout report. A pattern emerges which indicates user preference for learning tool may inform the participants' preference for animation in terms of peer recommendation, enjoyability, ease of understanding, and success at communicating complex information.

Figure 37 visualizes the fields of work/study for all participants. Figure 38 visualizes the fields of work/study broken down by preference for either whiteboard or 3D animation. 31% of participants who preferred the whiteboard animation are in the Biomedical Visualization program, but only 5% are in Pharmacy, 8% are in Medicine, 3% are in Nursing, and 8% are in Biological Sciences. For participants who preferred the 3D animation, however, 21% are in Biomedical Visualization, 10% are in Medicine, 13% are in Pharmacy, 3% are in Nursing, and 21% are in Biological Sciences. It appears that a greater proportion of students in Medicine, Pharmacy, and Biological Sciences prefer the 3D animation.

Figure 39 visualizes all participant responses to "How difficult was the content to understand?" from "Very easy" to "Very difficult." Figure 40 visualizes these responses broken down by preference for either whiteboard or 3D animation. It appears that a greater proportion of participants who responded "Very easy," "Easy," or "Neither easy nor hard" preferred the 3D animation over the whiteboard animation. The visualizations also show that a greater proportion of participants who responded "Difficult" or "Very difficult" preferred the whiteboard animation more.

Figures 41 – 43 convey both the total responses for and the breakdown of self-reported level of familiarity with bacterial signaling by preference for either whiteboard or 3D animation. It appears that a much greater proportion of participants who preferred the whiteboard animation were either “Not at all familiar” (40%) or “Slightly familiar” (40%) with bacterial signaling with respect to the participants who preferred the 3D animation. Reflecting past patterns of the breakout report, a larger proportion of those either “Extremely familiar” or “Moderately familiar” with bacterial signaling preferred the 3d animation over the whiteboard animation.

Figure 44 demonstrates both total and breakdown by preference responses to the question, “Which animation is more successful at communicating complex information? Although the total responses demonstrate a nearly 50/50 split, or rather, 47% whiteboard preference and 53% 3D preference, the breakout report shows a fascinating yet predictable pattern. 81% of participants who preferred the whiteboard animation as a learning tool thought that it was more successful at communicating complex information, and 90% of those who preferred the 3D animation as a learning tool thought that it was more successful. This pattern continues, as 86% of those who preferred the whiteboard animation thought that the whiteboard animation was easiest to understand, 60% thought that the whiteboard animation was the most enjoyable, and 76% would recommend the whiteboard animation to their peers.

#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	whiteboard animation	1.00	9.00	7.05	2.50	6.25	39
2	3D animation	1.00	9.00	5.62	2.70	7.31	39

#	Field	whiteboard animation		3D animation		Total
1	Medicine	33.33%	2	66.67%	4	6
2	Pharmacy	37.50%	3	62.50%	5	8
3	Nursing	50.00%	1	50.00%	1	2
4	Public Health	0.00%	0	100.00%	1	1
5	Biological Sciences	27.27%	3	72.73%	8	11
6	Chemical Sciences	33.33%	1	66.67%	2	3
7	Social Sciences	42.86%	3	57.14%	4	7
8	Biomedical Visualization	60.00%	12	40.00%	8	20
9	Other:	70.00%	14	30.00%	6	20

Figure 36. Breakdown of field of work/study by viewer preference for whiteboard animation or 3D animation

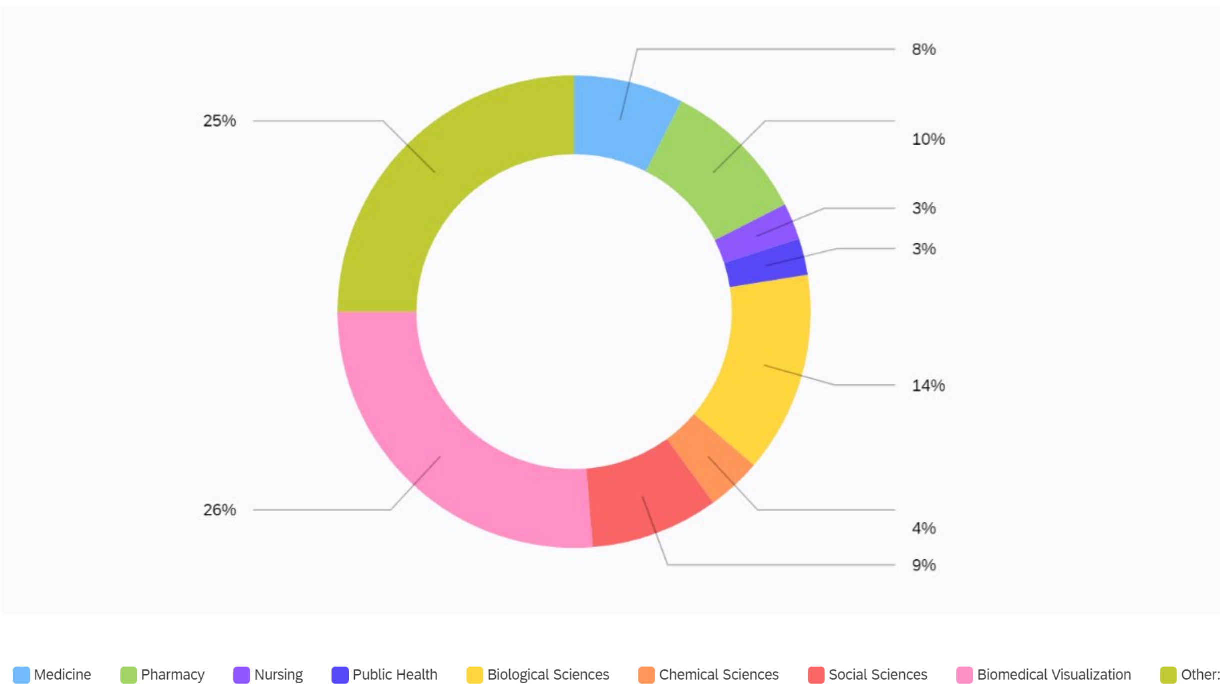


Figure 37. Visualization of participant field of work/study

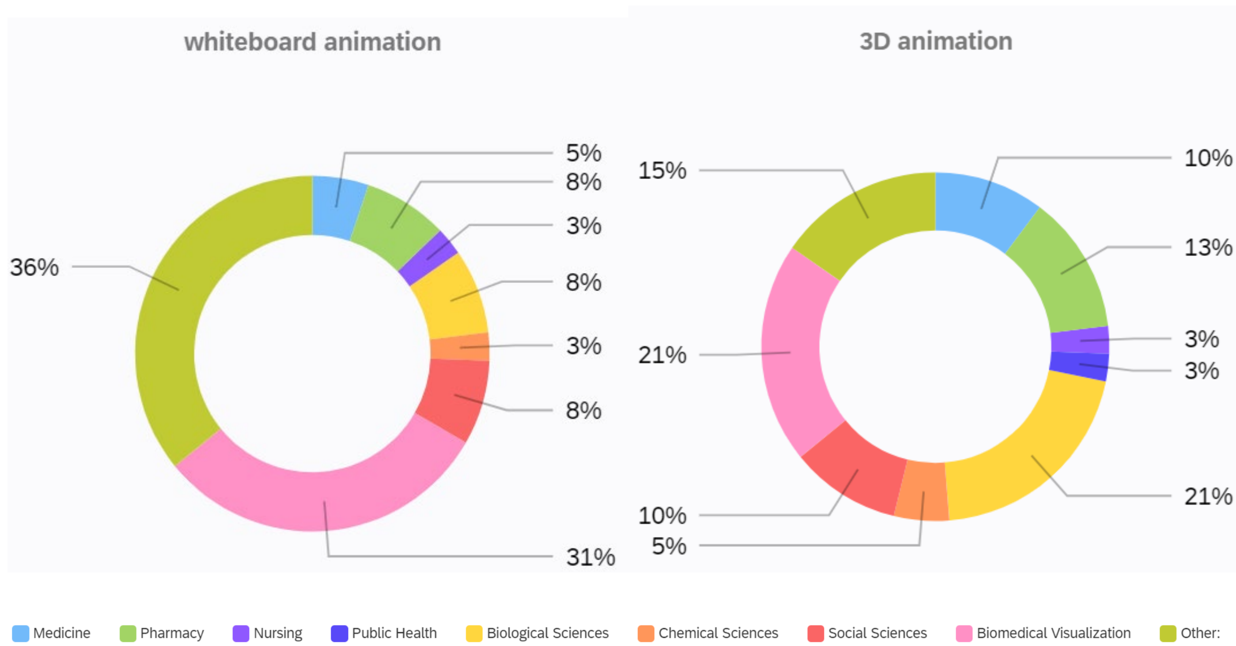


Figure 38. Visualization of field of work/study by viewer preference for whiteboard animation or 3D animation

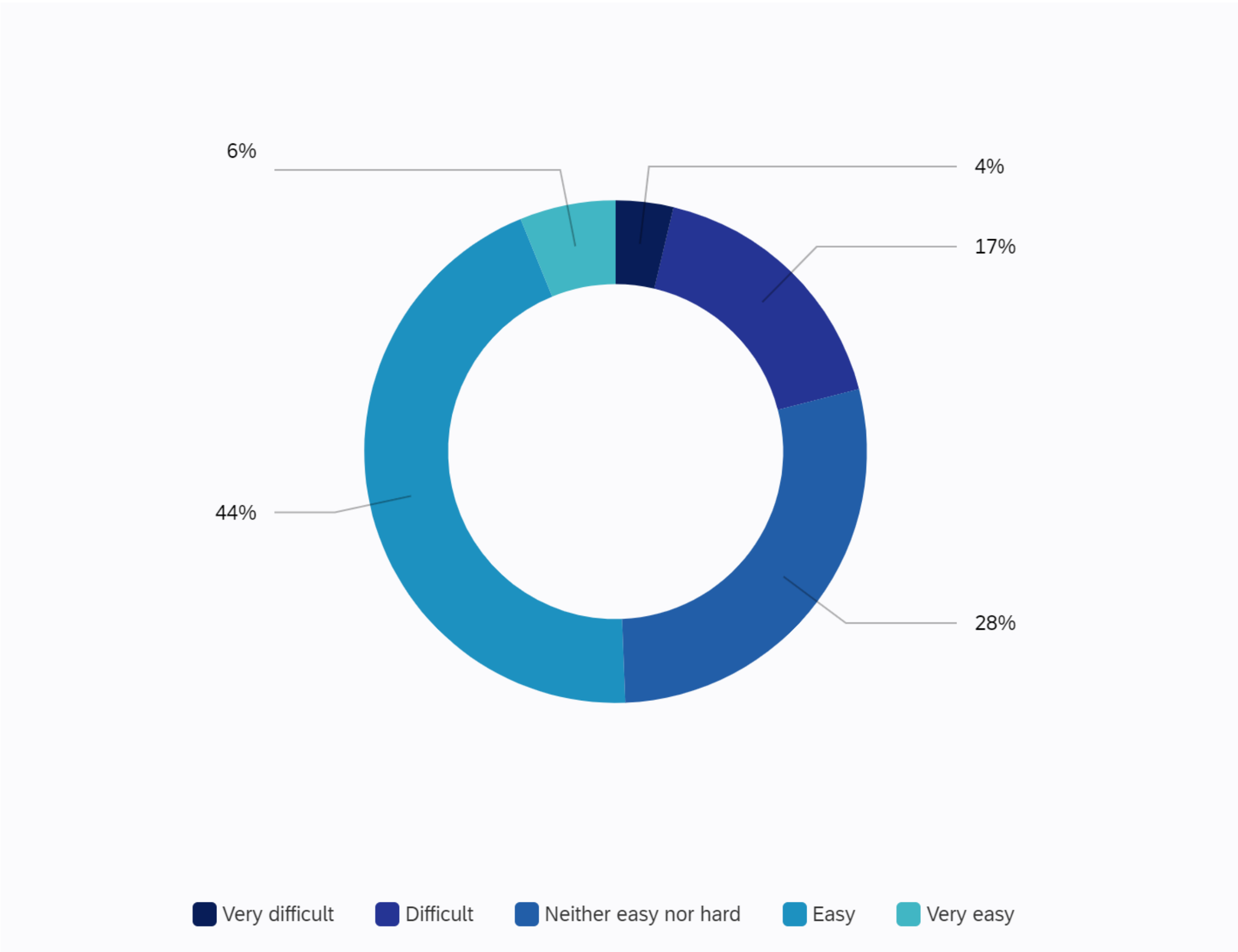


Figure 39. How difficult was the content to understand?

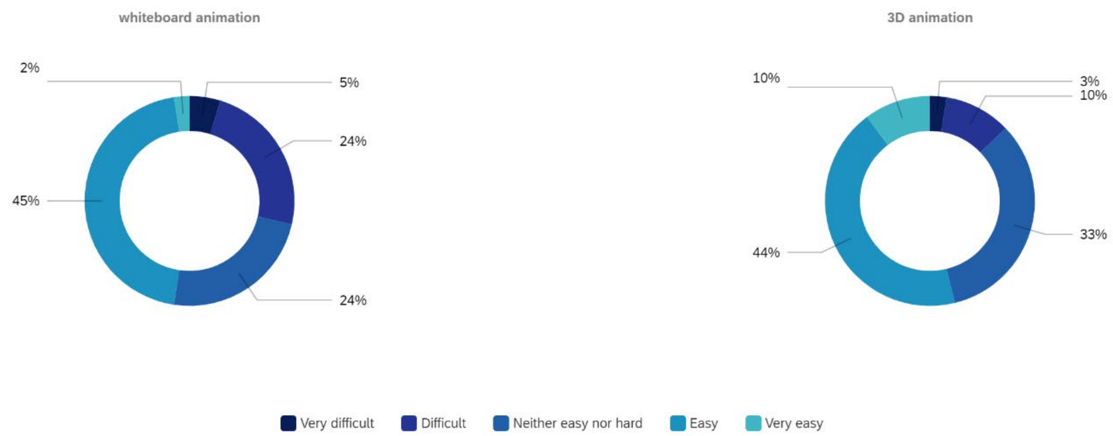


Figure 40. Viewer preference breakdown of responses to: "How difficult was the content to understand?"

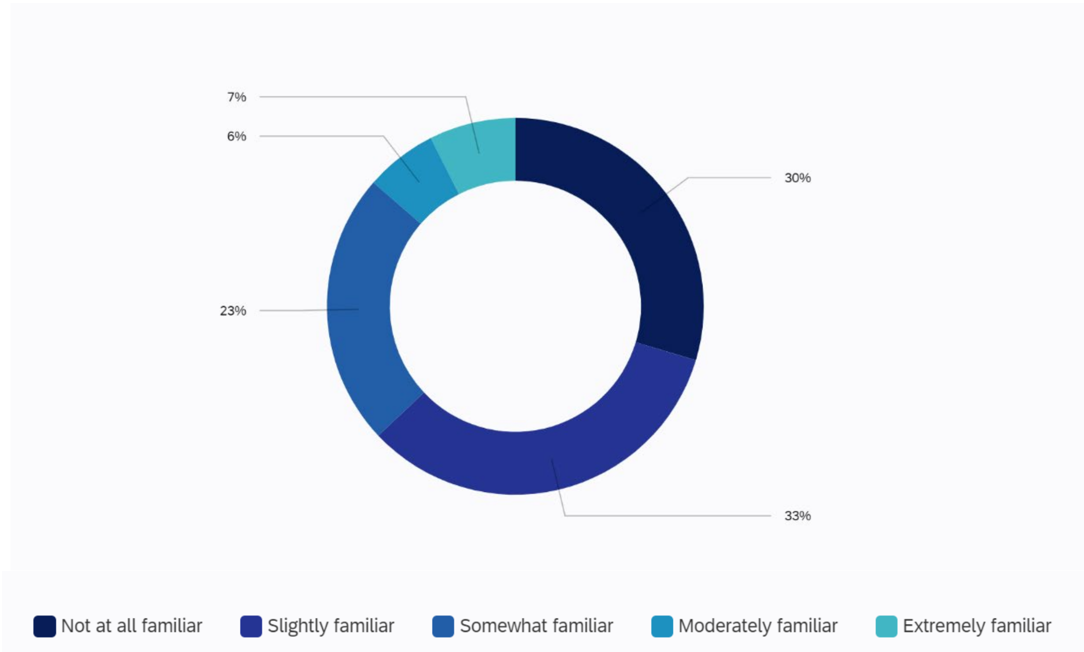
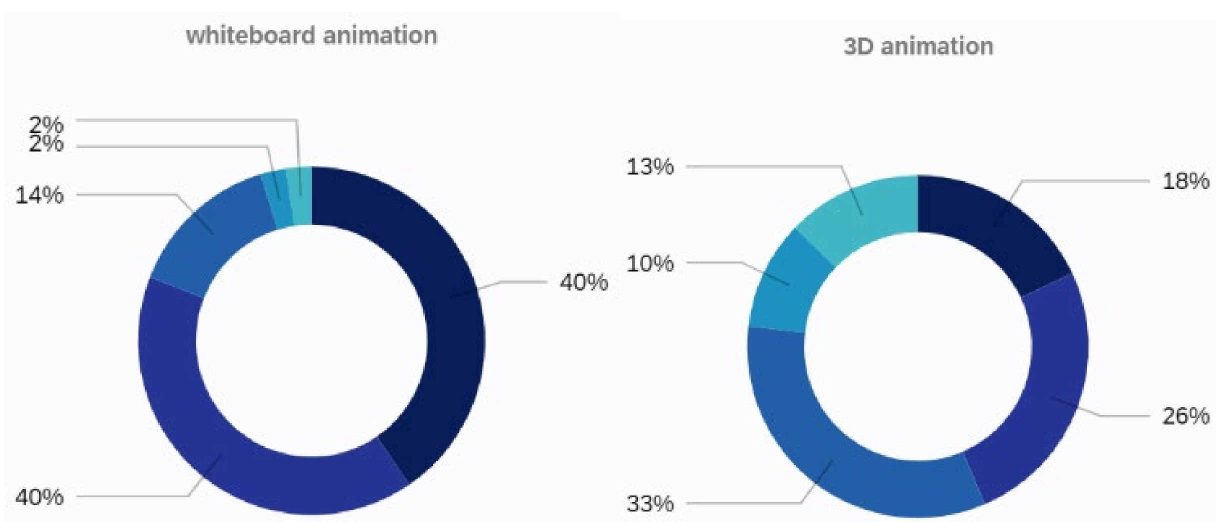


Figure 41. Visualization of total participant response to: Rate your level of knowledge with bacterial signaling.

#	Field	whiteboard animation		3D animation	
3	Somewhat familiar	14.29%	6	33.33%	13
2	Slightly familiar	40.48%	17	25.64%	10
1	Not at all familiar	40.48%	17	17.95%	7
5	Extremely familiar	2.38%	1	12.82%	5
4	Moderately familiar	2.38%	1	10.26%	4
			42		39

Figure 42. Preference breakdown of level of familiarity with bacterial signaling



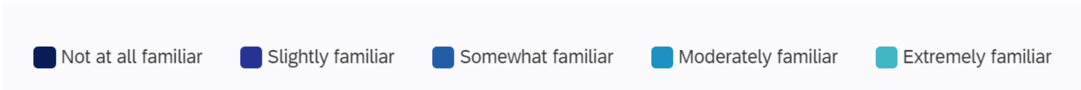
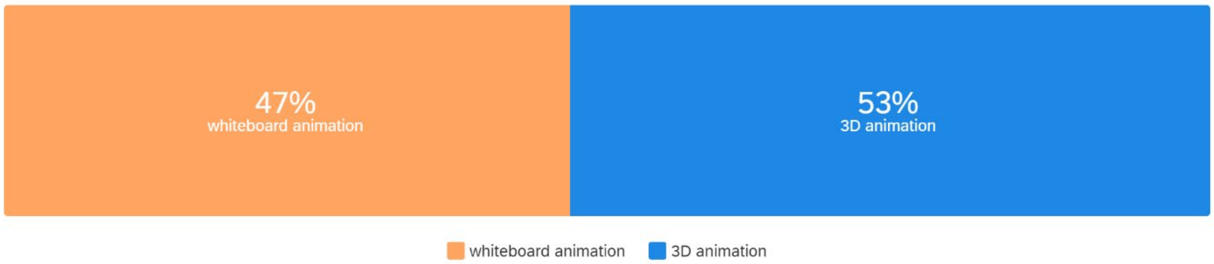


Figure 43. Visualization of reference breakdown of level of familiarity with bacterial signaling



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	whiteboard animation	1.00	2.00	1.19	0.39	0.15	42
2	3D animation	1.00	2.00	1.90	0.30	0.09	39

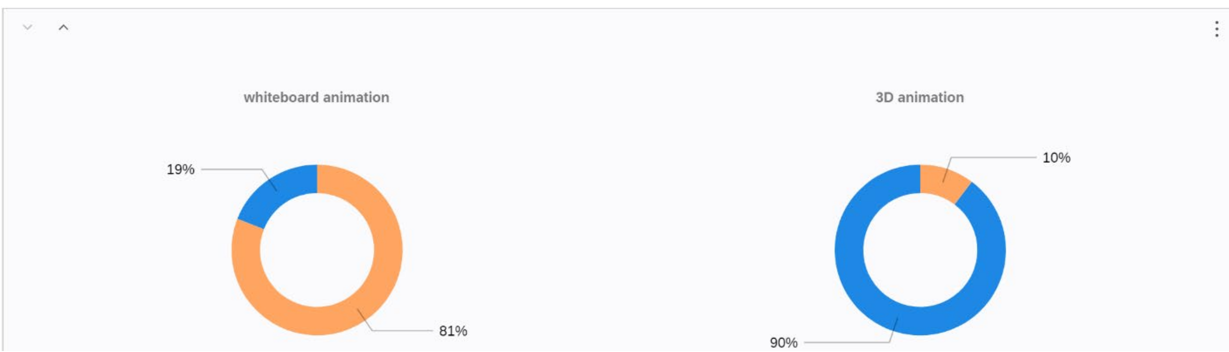
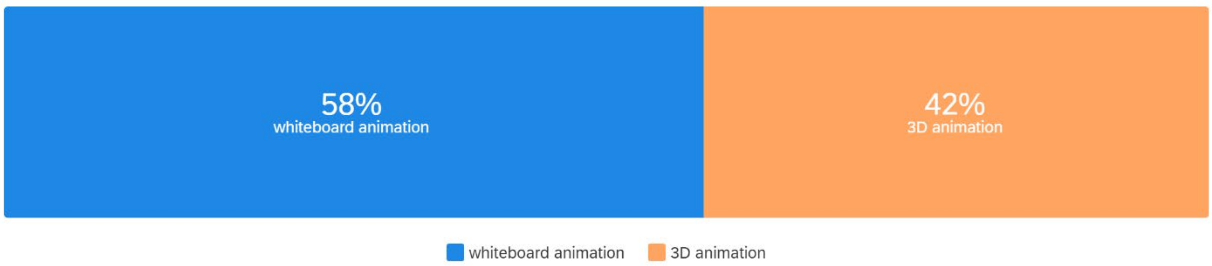


Figure 44. Breakdown of which animation is more successful at communicating complex information by viewer preference



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	whiteboard animation	1.00	2.00	1.14	0.35	0.12	42
2	3D animation	1.00	2.00	1.72	0.45	0.20	39

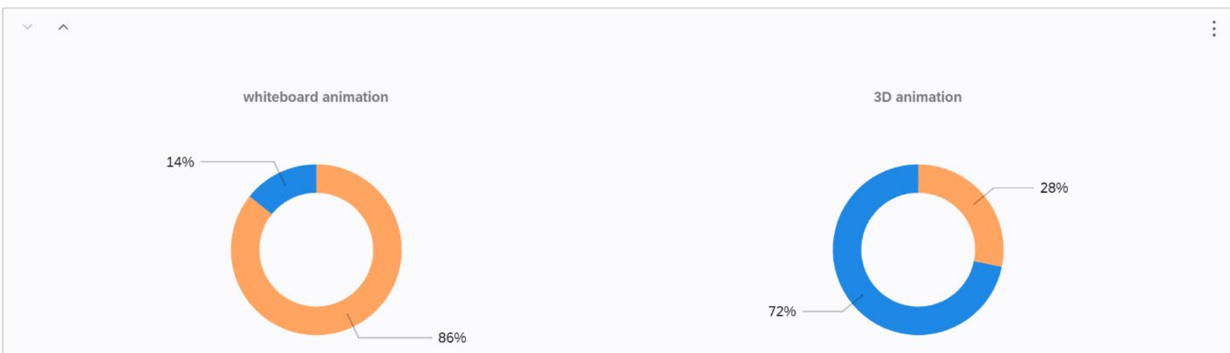
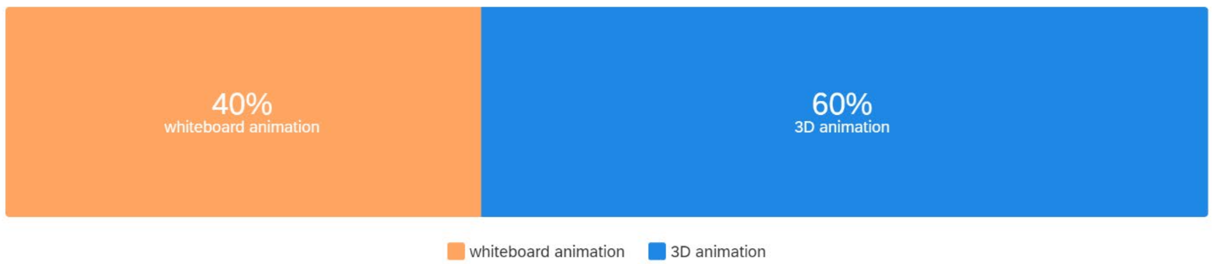
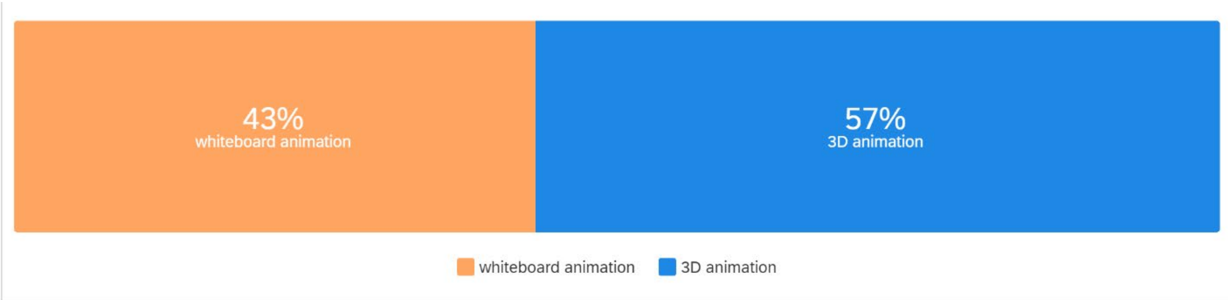


Figure 45. Breakdown of which animation is easiest to understand by viewer preference



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	whiteboard animation	1.00	2.00	1.40	0.49	0.24	42
2	3D animation	1.00	2.00	1.82	0.38	0.15	39

Figure 46. Breakdown of which animation was most enjoyable by viewer preference



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	whiteboard animation	1.00	2.00	1.24	0.43	0.18	42
2	3D animation	1.00	2.00	1.92	0.27	0.07	39

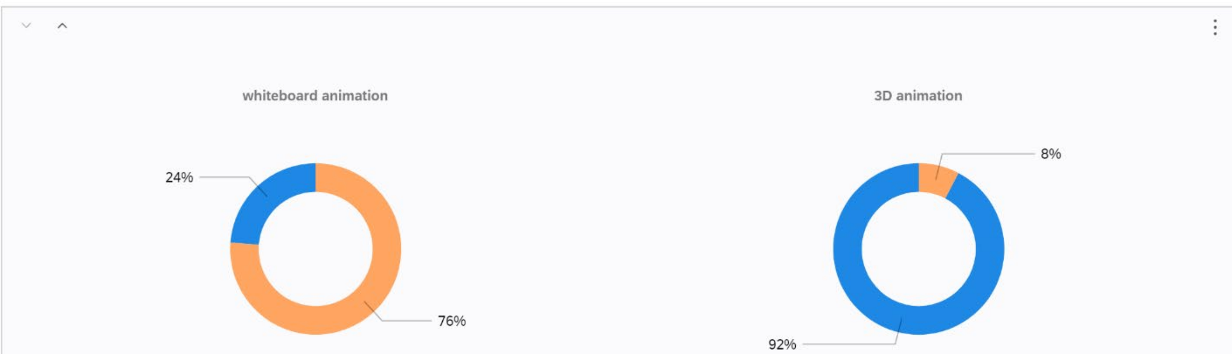


Figure 47. Breakdown of which animation participants would recommend to peers by viewer preference

I. <u>Breakdown by familiarity with bacterial signaling</u>

In order to determine how viewer preference changed with the level of prior knowledge of bacterial signaling, UIC Qualtrics' Breakout Report feature was used. Figures 48 to 53 illustrate this data analysis. It appeared that participants with higher levels of prior knowledge with bacterial signaling were more likely to select the 3D animation as a learning tool, characterize it as more enjoyable, easier to understand, and more successful at communicating complex information, and recommend it to peers. The opposite trend occurred in participants with low familiarity. These participants overwhelmingly preferred the whiteboard animation. 83% of participants “Extremely familiar” and 80% of participants “Moderately familiar” with bacterial signaling preferred the 3D animation as a learning tool. However, 71% of participants “Not at all familiar” and 63% of participants “Slightly familiar” preferred to use the whiteboard animation as a learning tool. 83% of participants “Not at all familiar” with bacterial signaling found the whiteboard animation easiest to understand and 71% thought that it was more successful at communicating complex information.

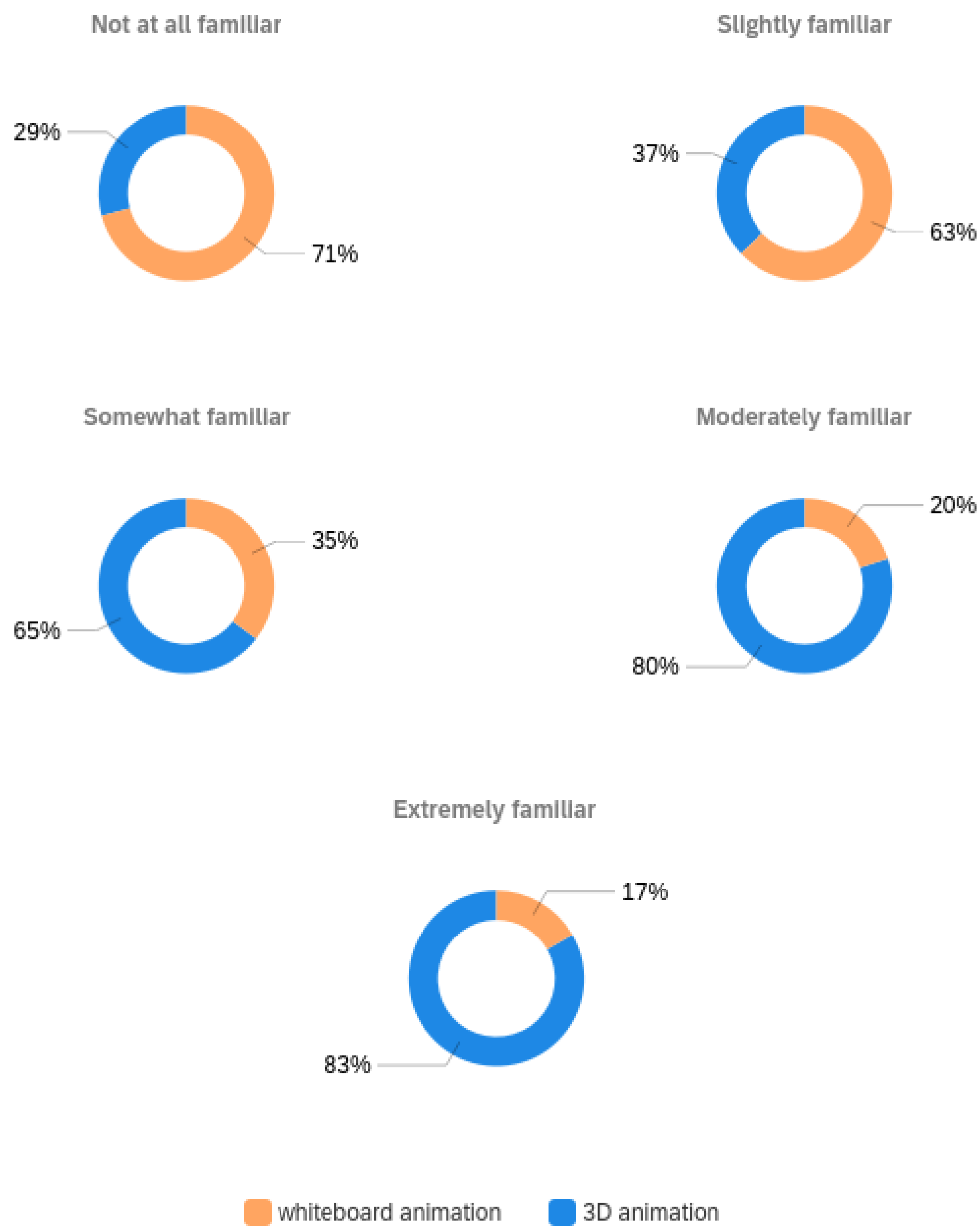


Figure 48. Breakdown of viewer preference for learning tool by familiarity with bacterial signaling

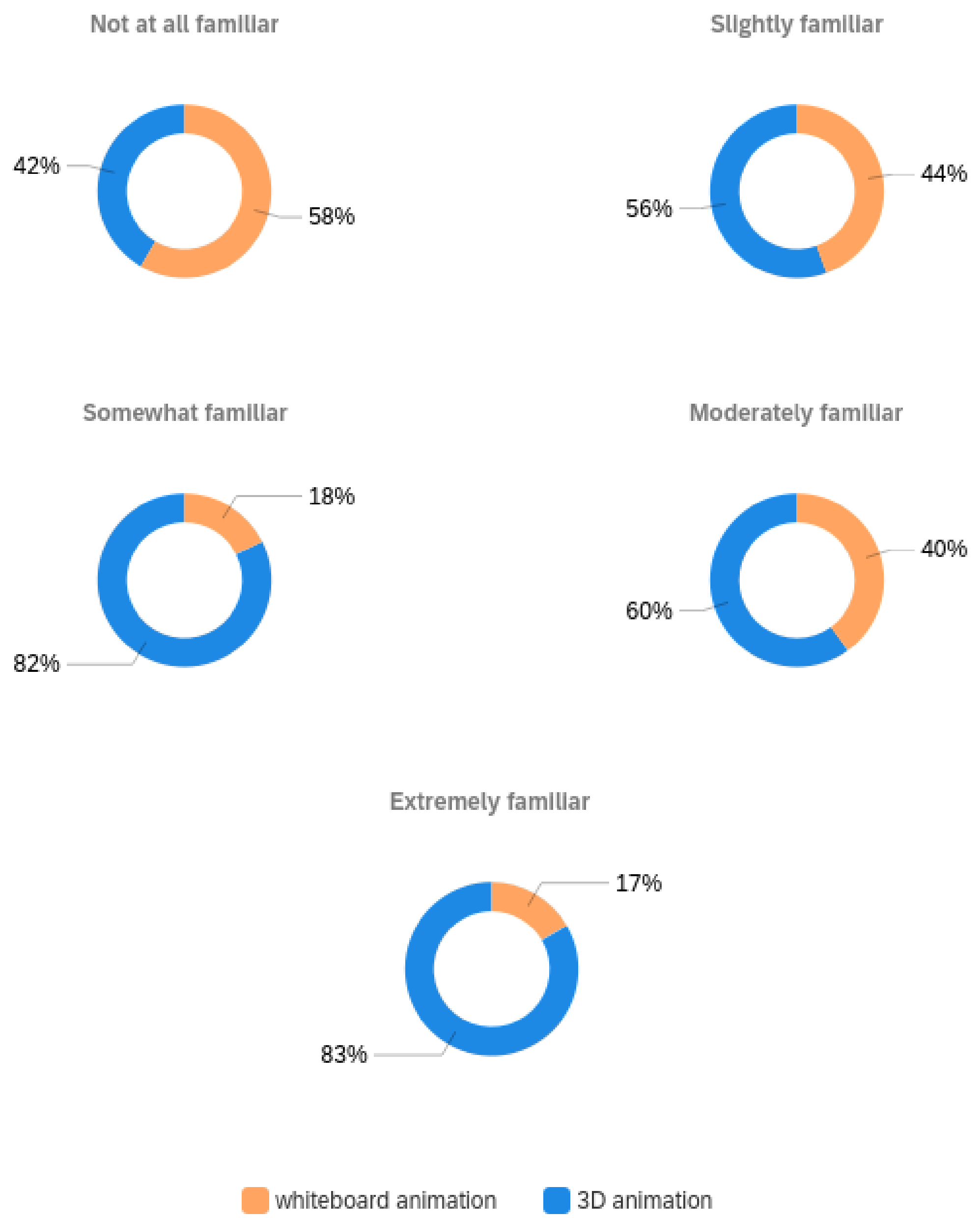


Figure 49. Breakdown of most enjoyable animation by familiarity with bacterial signaling

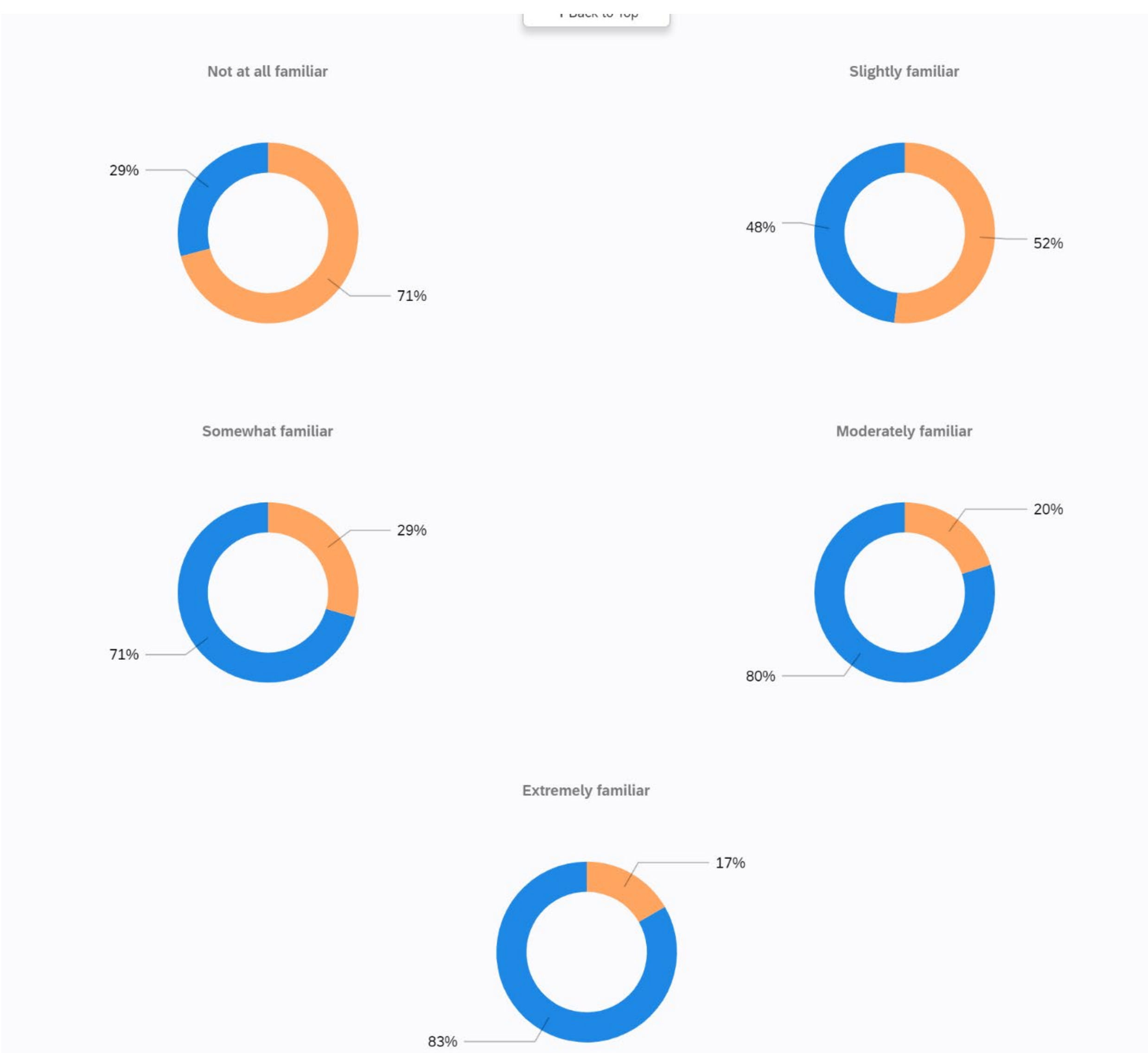


Figure 50. Breakdown of which animation is more successful at communicating complex information by familiarity with bacterial signaling

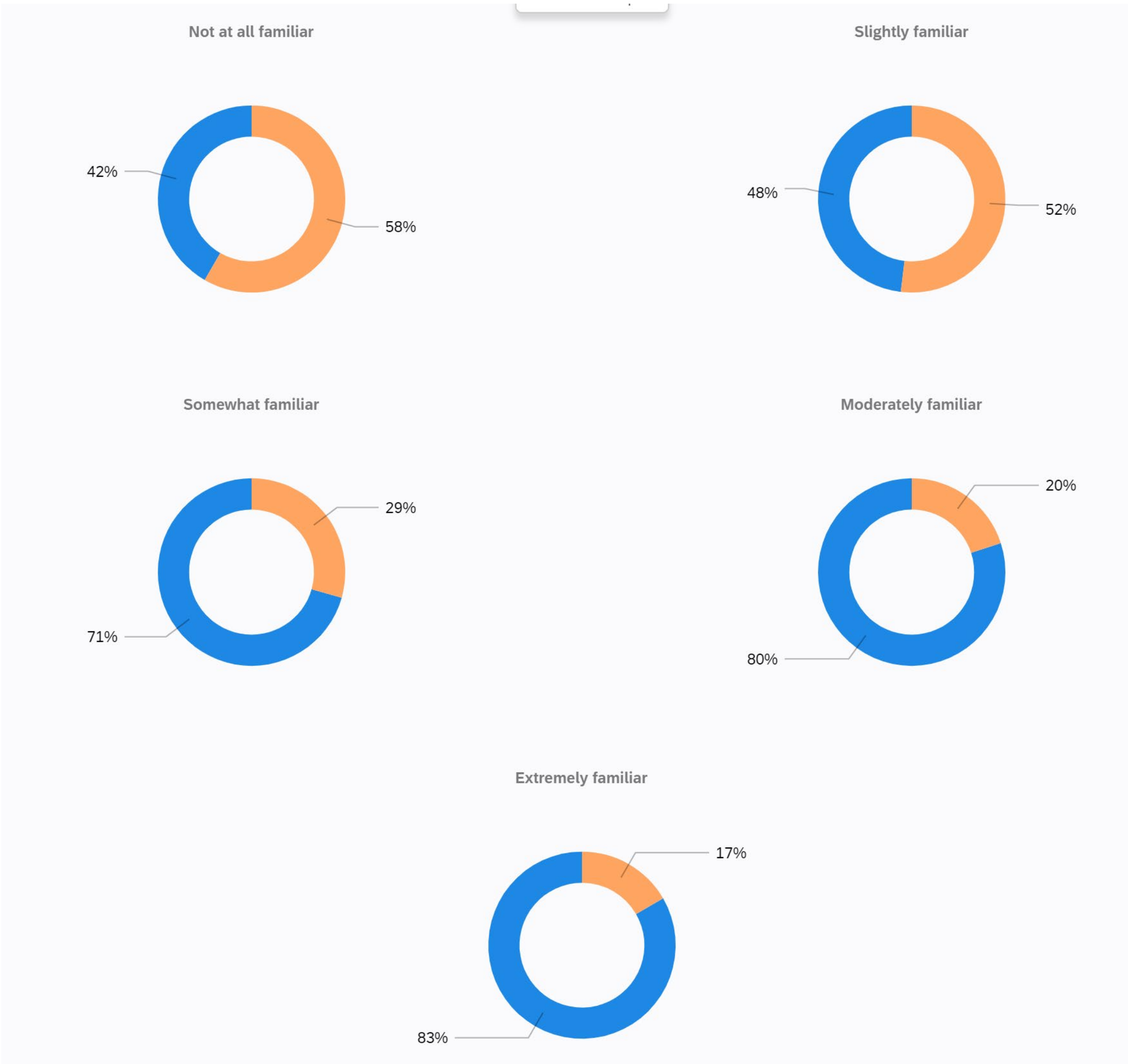


Figure 51. Breakdown of which animation participants would most likely recommend to peers by familiarity with bacterial signaling

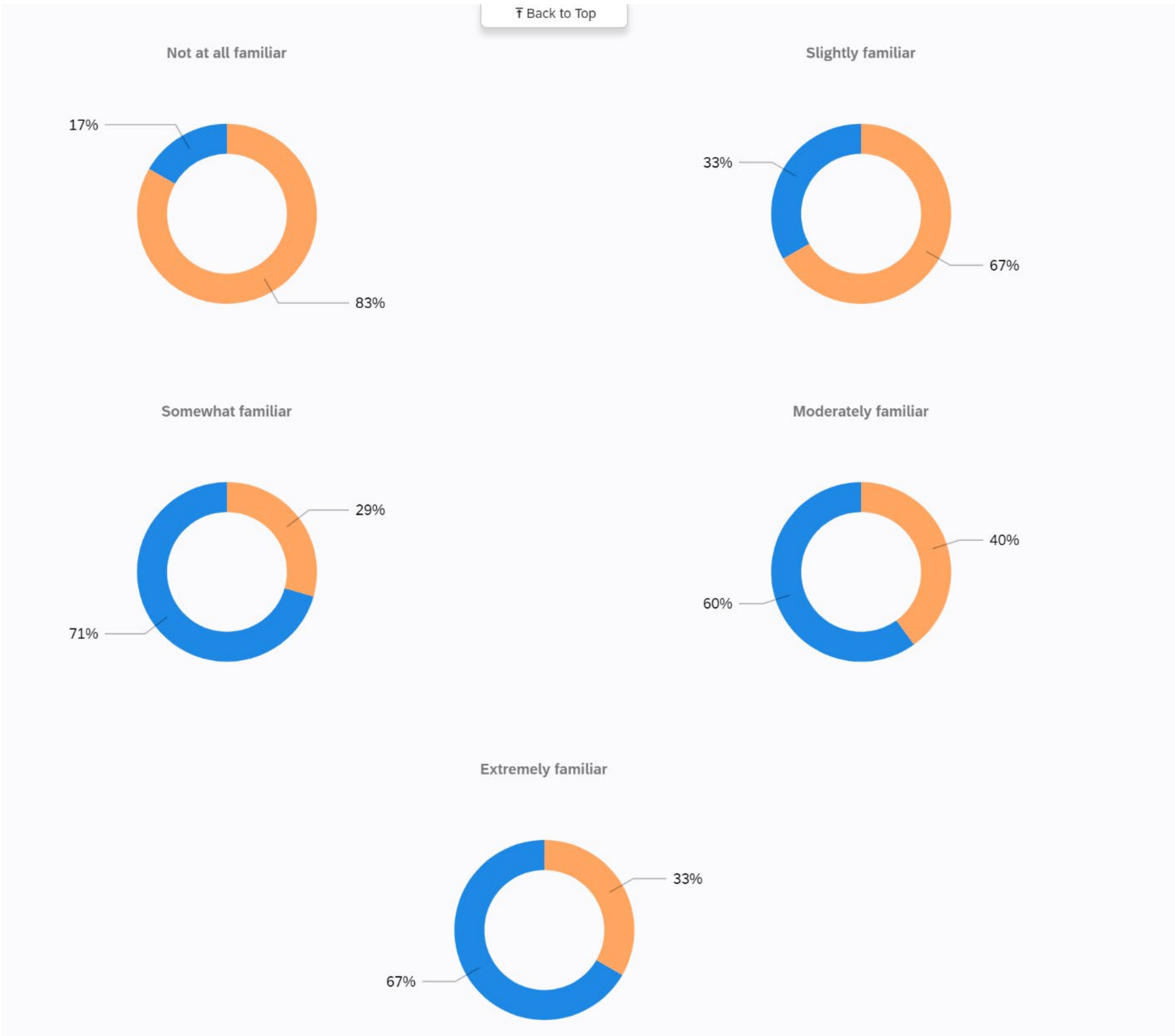
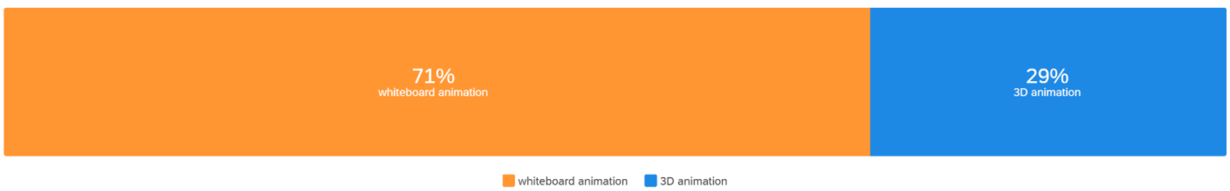


Figure 52. Breakdown of which animation was easiest to understand by familiarity with bacterial signaling

J. <u>Characteristics of participants with low level of prior knowledge of bacterial signaling</u>

To observe the preferences of participants with the lowest level of prior knowledge of bacterial signaling, UIC Qualtrics was used to filter all participants who responded that they were “Not at all familiar” with bacterial signaling. Visualizations of this report are shown in Figures 54 to 58. This group demonstrated an overwhelming preference for whiteboard animation as a learning tool, thought that the whiteboard animation was the easiest and most enjoyable animation to understand, was more successful at communicating complex information, and would more likely be recommended by them to peers.

Q54 - I prefer to use the ________ as a learning tool. Page Options

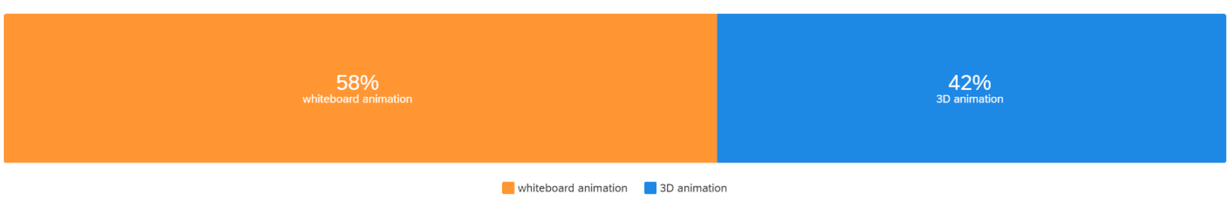


#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	I prefer to use the ________ as a learning tool.	1.00	2.00	1.29	0.45	0.21	24

#	Field		Choice Count
1	whiteboard animation	70.83%	17
2	3D animation	29.17%	7
			24

Figure 53. Low familiarity participants' learning tool preference

Q55 - I enjoyed the __________ most. Page Options



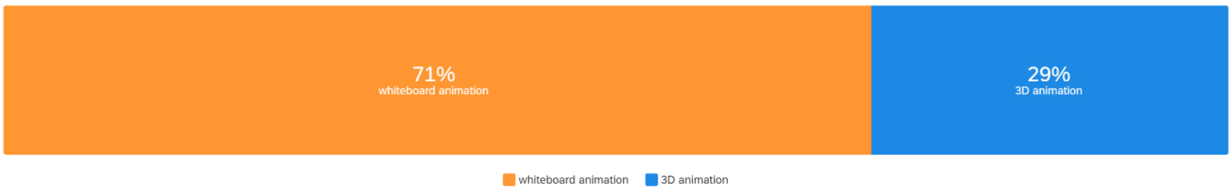
#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	I enjoyed the ________ most.	1.00	2.00	1.42	0.49	0.24	24

#	Field		Choice Count
1	whiteboard animation	58.33%	14
2	3D animation	41.67%	10
			24

Figure 54. Animation enjoyed the most by low familiarity participants

Q56 - The _________ is more successful at communicating complex information.

Page Options



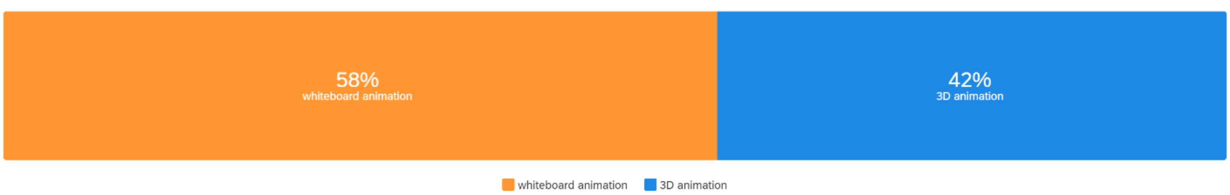
#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	The ________ is more successful at communicating complex information.	1.00	2.00	1.29	0.45	0.21	24

#	Field		Choice Count
1	whiteboard animation	70.83%	17
2	3D animation	29.17%	7
			24

Figure 55. Low familiarity participants' preference of which animation is more successful at communicating complex information

Q57 - I am more likely to recommend the __________ to peers.

Page Options

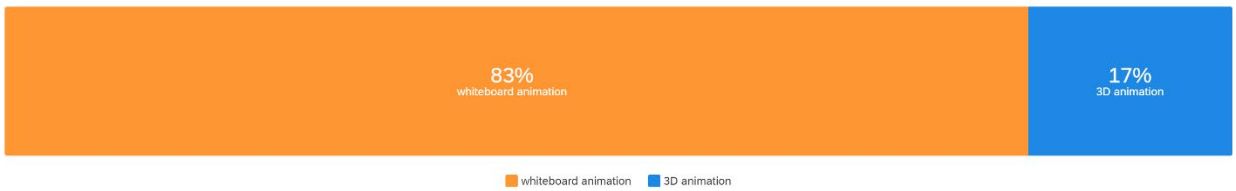


#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	I am more likely to recommend the _________ to peers.	1.00	2.00	1.42	0.49	0.24	24

#	Field		Choice Count
1	whiteboard animation	58.33%	14
2	3D animation	41.67%	10
			24

Figure 56. Animation most likely to be recommend to peers by low familiarity participants

Q58 - The ______ was easiest to understand. Page Options



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	The _____ was easiest to understand.	1.00	2.00	1.17	0.37	0.14	24

#	Field	Choice Count	
1	whiteboard animation	83.33%	20
2	3D animation	16.67%	4
			24

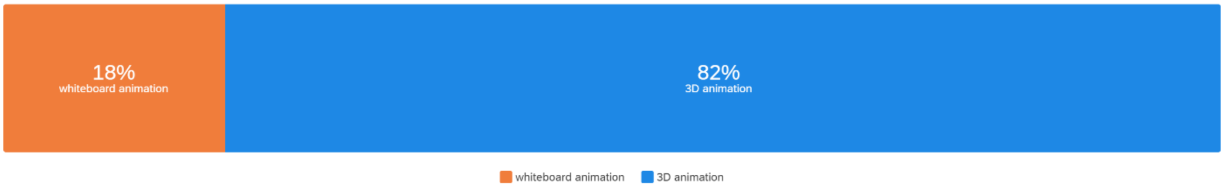
Figure 57. Animation easiest to understand for low familiarity participants

K. <u>Characteristics of participants with high level of prior knowledge of bacterial signaling</u>

To observe the preferences of participants with the highest level of prior knowledge of bacterial signaling, UIC Qualtrics was used to filter all participants who responded that they were “Extremely familiar” or “Moderately familiar” with bacterial signaling. Visualizations of this report are shown in Figures 59 to 63. This group demonstrated an overwhelming preference (82% of responses) for 3D animation as a learning tool. Participants with relatively high levels of prior knowledge thought that the 3D animation was the easiest and most enjoyable animation to understand, was more successful at communicating complex information, and would more likely be recommended by them to peers.

Q54 - I prefer to use the ________ as a learning tool.

Page Options



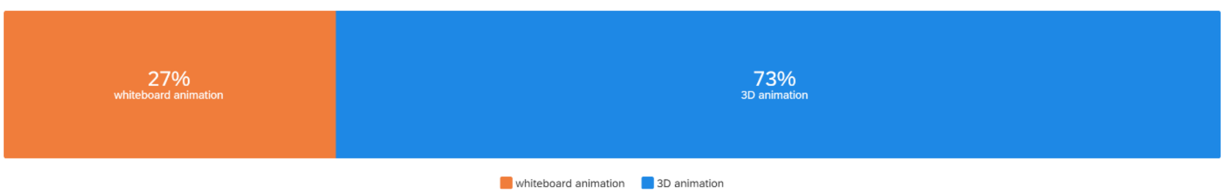
#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	I prefer to use the ________ as a learning tool.	1.00	2.00	1.82	0.39	0.15	11

#	Field	Choice Count	
1	whiteboard animation	18.18%	2
2	3D animation	81.82%	9
			11

Figure 58. High familiarity participants' learning tool preference

Q55 - I enjoyed the __________ most.

Page Options



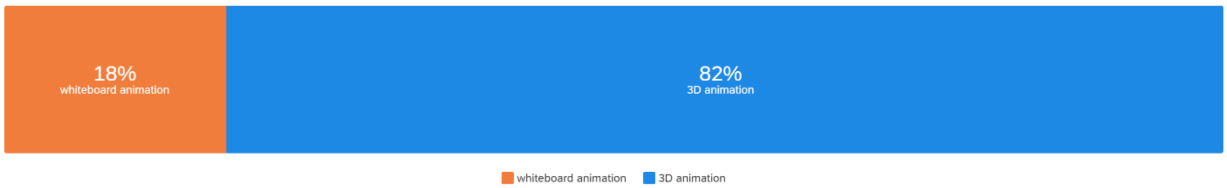
#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	I enjoyed the __________ most.	1.00	2.00	1.73	0.45	0.20	11

#	Field	Choice Count	
1	whiteboard animation	27.27%	3
2	3D animation	72.73%	8
			11

Figure 59. Animation enjoyed the most by high familiarity participants

Q56 - The _________ is more successful at communicating complex information.

Page Options



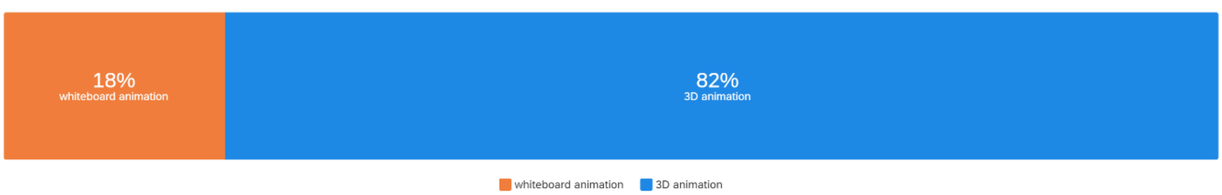
#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	The ________ is more successful at communicating complex information.	1.00	2.00	1.82	0.39	0.15	11

#	Field		Choice Count
1	whiteboard animation	18.18%	2
2	3D animation	81.82%	9
			11

Figure 60. High familiarity participants' preference of which animation is more successful at communicating complex information

Q57 - I am more likely to recommend the __________ to peers.

Page Options



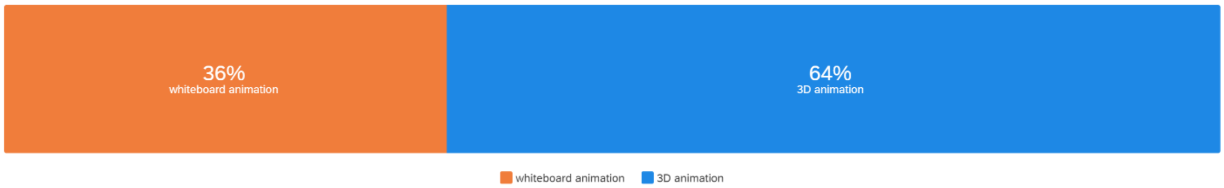
#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	I am more likely to recommend the _________ to peers.	1.00	2.00	1.82	0.39	0.15	11

#	Field		Choice Count
1	whiteboard animation	18.18%	2
2	3D animation	81.82%	9
			11

Figure 61. Figure 57. Animation most likely to be recommend to peers by high familiarity participants

Q58 - The _____ was easiest to understand.

Page Options



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	The _____ was easiest to understand.	1.00	2.00	1.64	0.48	0.23	11

#	Field		Choice Count
1	whiteboard animation	36.36%	4
2	3D animation	63.64%	7
			11

Figure 62. Animation easiest to understand for high familiarity participants

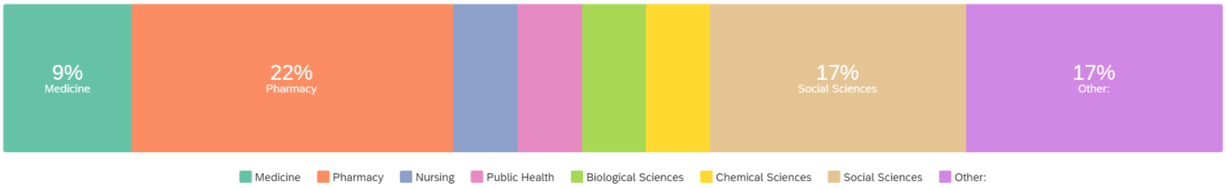
L. <u>Characteristics of participants with low spatial reasoning</u>

UIC Qualtrics was used to generate a report of all participants with low spatial reasoning, or those who got 3 questions or less correct on the spatial reasoning test. This number was chosen because the mean score on the spatial reasoning test was 4/7. Figure 63 shows the fields of work/study for this group, most of which are aggregated in Pharmacy. Many low spatial-reasoning participants are in Medicine or Social Sciences, and 17% responded as “Other.” Low spatial reasoning participants also gravitated towards little to no prior knowledge of bacterial signaling: 21% are not at all familiar, 25% are slightly familiar, and 29% are somewhat familiar (Figure 64).

Next, the low spatial reasoning participants’ perceptions of the 3D animation was analyzed, as shown in Figures 65 - 69. 17% found the 3D animation “Somewhat useless” and 21% found it “Slightly useful.” However, 50% found it either “Slightly useful” or “Somewhat useful.” 38% of low spatial reasoning participants agreed that they enjoyed the 3D quality of the animation and that the information presented in the animation was moderately clear. Figures 70 - 74 demonstrate low spatial reasoning participants’ perceptions of the whiteboard animation. 25% of these participants found the whiteboard animation “Somewhat useful” and 25% found it “Somewhat useless” for bacterial electrochemical signaling. 46% agreed that they enjoyed the 2D quality and 35% somewhat agreed that they enjoyed the simplicity of the whiteboard animation. Just like the 3D animation, 38% of low spatial reasoning participants found the whiteboard animation to be moderately clear.

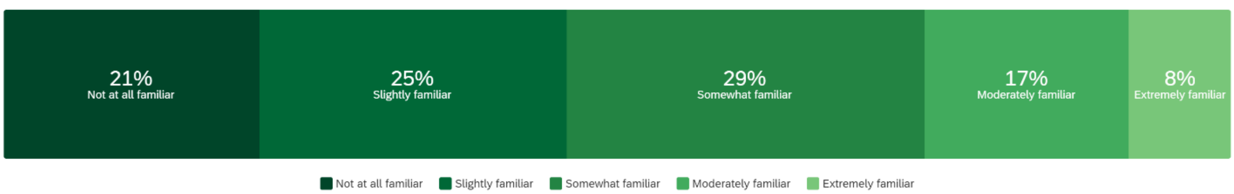
The results of low spatial reasoning participant responses to the preference survey was quite surprising. Although it was expected that this group would gravitate towards preference of the whiteboard animation for learning, ease of use, enjoyability, recommendation to peers, and communicating complex concepts, this was not so. Only 42% preferred to use the whiteboard animation as a learning tool and were likely to recommend it to peers. The participants split evenly 50/50 between which animation they enjoyed the most and which animation they thought was more successful at communicating complex information. Lastly, 46% of low spatial reasoning participants thought that the whiteboard animation was easiest to understand.

Figures 80 – 82 visualize the self-reported cognitive load survey, or perceived mental difficulty survey, taken by low spatial reasoning participants. Although 42% of participants found the content easy to understand, 38% of participants required some effort to comprehend the content. Additionally, 38% of participants were only slightly familiar with bacterial signaling.



#	Field		Choice Count
1	Medicine	8.70%	2
2	Pharmacy	21.74%	5
3	Nursing	4.35%	1
4	Public Health	4.35%	1
5	Biological Sciences	4.35%	1
6	Chemical Sciences	4.35%	1
7	Social Sciences	17.39%	4
8	Biomedical Visualization	17.39%	4
9	Other:	17.39%	4
			23

Figure 63. Fields of work/study for participants with low spatial reasoning



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	Rate your level of prior knowledge with bacterial signaling.	2.00	5.00	3.27	1.21	1.47	11

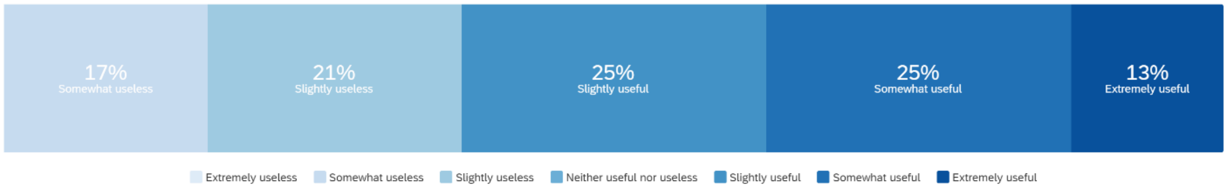
Retrieving data...

#	Field		Choice Count
1	Not at all familiar	0.00%	0
2	Slightly familiar	36.36%	4
3	Somewhat familiar	27.27%	3
4	Moderately familiar	9.09%	1
5	Extremely familiar	27.27%	3
			11

Figure 64. Prior knowledge of bacterial signaling for participants with low spatial reasoning

Q41 - How useful is this animation for understanding bacterial electrochemical signaling?

Page Options

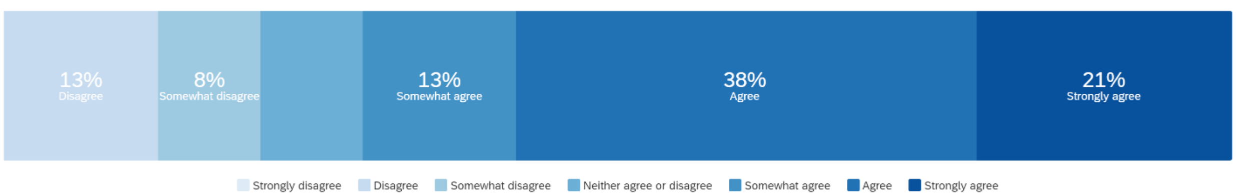


#	Field	Choice Count	
2	Somewhat useless	16.67%	4
6	Somewhat useful	25.00%	6
3	Slightly useless	20.83%	5
5	Slightly useful	25.00%	6
4	Neither useful nor useless	0.00%	0
1	Extremely useless	0.00%	0
7	Extremely useful	12.50%	3
			24

Figure 65. Low spatial reasoning participant response to: How useful is this [3D] animation for understanding bacterial electrochemical signaling?

Q42 - How strongly do you agree or disagree with this statement: I enjoyed the 3D quality of the animation.

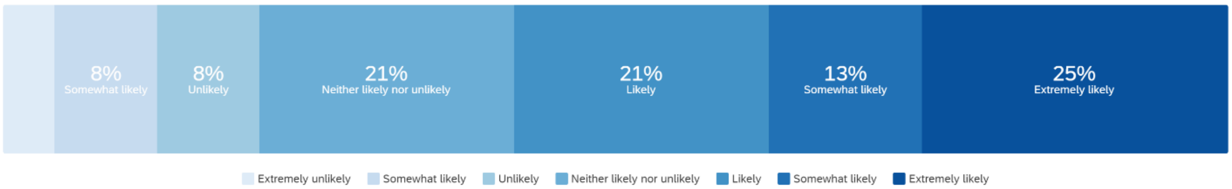
Page Options



#	Field	Choice Count	
1	Strongly disagree	0.00%	0
7	Strongly agree	20.83%	5
3	Somewhat disagree	8.33%	2
5	Somewhat agree	12.50%	3
4	Neither agree or disagree	8.33%	2
2	Disagree	12.50%	3
6	Agree	37.50%	9
			24

Figure 66. Low spatial reasoning participant response to: I enjoyed the 3D quality of this animation.

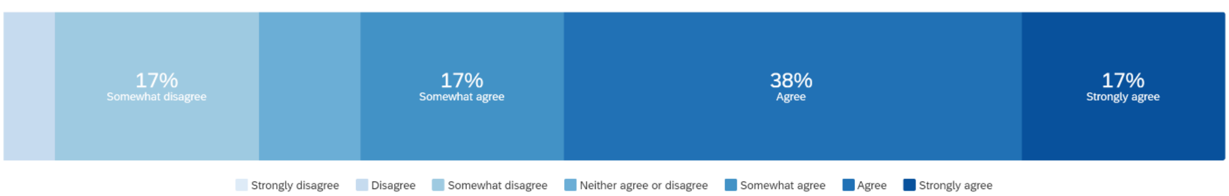
Q44 - How likely are you to use 3D animation to communicate your research? Page Options



#	Field		Choice Count
3	Unlikely	8.33%	2
6	Somewhat likely	12.50%	3
2	Somewhat likely	8.33%	2
4	Neither likely nor unlikely	20.83%	5
5	Likely	20.83%	5
1	Extremely unlikely	4.17%	1
7	Extremely likely	25.00%	6
			24

Figure 67. Low spatial reasoning participant response to: How likely are you to use 3D animation to communicate your research?

Q43 - How strongly do you agree or disagree with this statement: I enjoyed the complexity of the 3D animation. Page Options



#	Field		Choice Count
1	Strongly disagree	0.00%	0
2	Disagree	4.17%	1
3	Somewhat disagree	16.67%	4
4	Neither agree or disagree	8.33%	2
5	Somewhat agree	16.67%	4
6	Agree	37.50%	9
7	Strongly agree	16.67%	4
			24

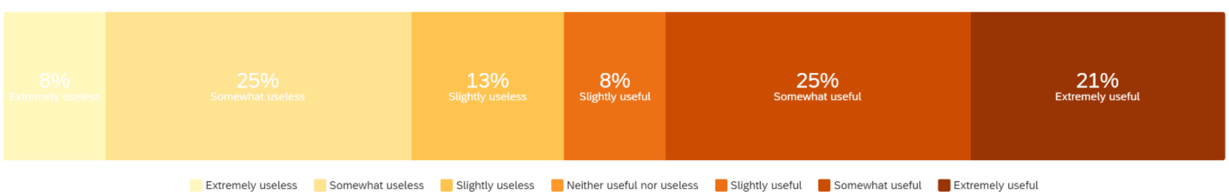
Figure 68. Low spatial reasoning participant response to: I enjoyed the complexity of the 3D animation

Q45 - How clear was the information presented in this animation? Page Options

#	Field		Choice Count
1	Extremely clear	20.83%	5
2	Moderately clear	37.50%	9
3	Slightly clear	16.67%	4
4	Neither clear nor unclear	8.33%	2
5	Slightly unclear	4.17%	1
6	Moderately unclear	8.33%	2
7	Extremely unclear	4.17%	1
			24

Figure 69. Low spatial reasoning participant response to: How clear was the information presented in this [3D] animation?

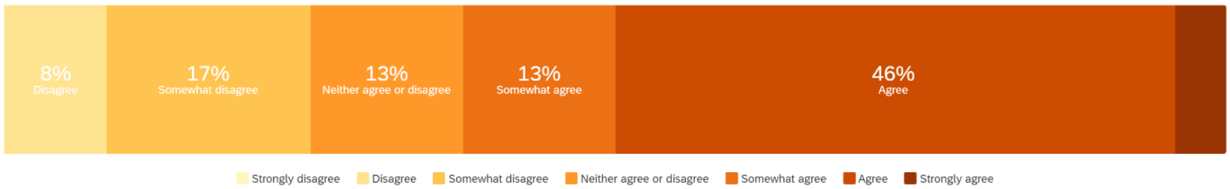
Q46 - How useful is this animation for understanding bacterial electrochemical signaling? Page Options



#	Field		Choice Count
1	Extremely useless	8.33%	2
2	Somewhat useless	25.00%	6
3	Slightly useless	12.50%	3
4	Neither useful nor useless	0.00%	0
5	Slightly useful	8.33%	2
6	Somewhat useful	25.00%	6
7	Extremely useful	20.83%	5
			24

Figure 70. Low spatial reasoning participant response to: How useful is this [whiteboard] animation for understanding bacterial electrochemical signaling?

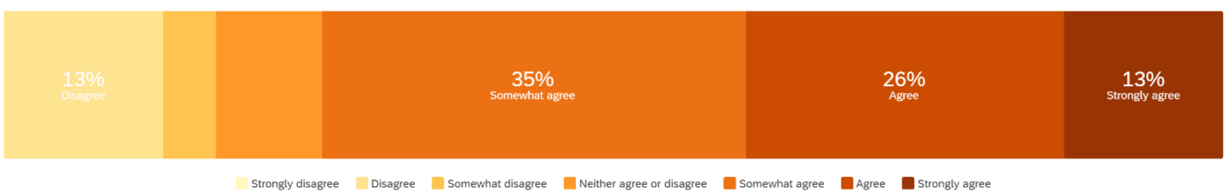
Q47 - How strongly do you agree or disagree with this statement: I enjoyed the 2D quality of the whiteboard animation. Page Options



#	Field		Choice Count
1	Strongly disagree	0.00%	0
7	Strongly agree	4.17%	1
3	Somewhat disagree	16.67%	4
5	Somewhat agree	12.50%	3
4	Neither agree or disagree	12.50%	3
2	Disagree	8.33%	2
6	Agree	45.83%	11
			24

Figure 71. Low spatial reasoning participant response to: I enjoyed the 2D quality of this animation

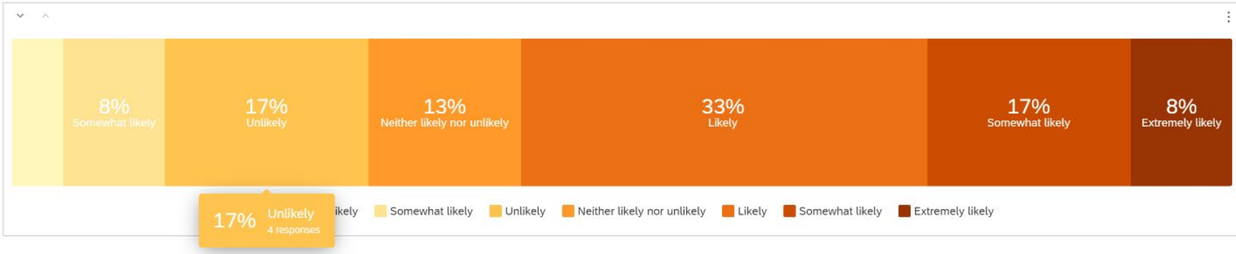
Q67 - How strongly do you agree or disagree with this statement: I enjoyed the simplicity of the 2D animation. Page Options



#	Field		Choice Count
1	Strongly disagree	0.00%	0
2	Disagree	13.04%	3
3	Somewhat disagree	4.35%	1
4	Neither agree or disagree	8.70%	2
5	Somewhat agree	34.78%	8
6	Agree	26.09%	6
7	Strongly agree	13.04%	3
			23

Figure 72. Low spatial reasoning participant response to: I enjoyed the simplicity of the 2D animation

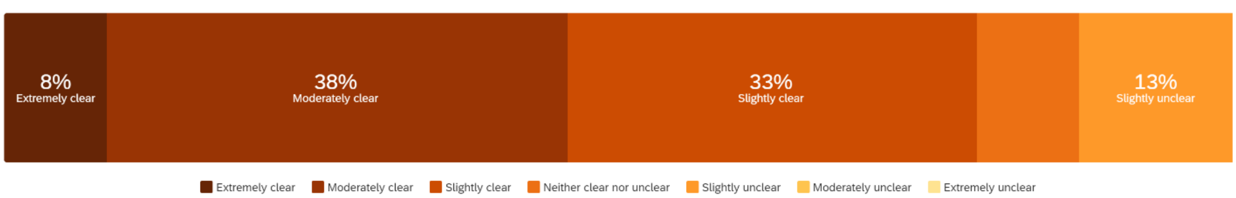
Q49 - How likely are you to use whiteboard animation to communicate your research? Page Options



#	Field		Choice Count
1	Extremely unlikely	4.17%	1
2	Somewhat likely	8.33%	2
3	Unlikely	16.67%	4
4	Neither likely nor unlikely	12.50%	3
5	Likely	33.33%	8
6	Somewhat likely	16.67%	4
7	Extremely likely	8.33%	2
			24

Figure 73. Low spatial reasoning participant response to: How likely are you to use whiteboard animation to communicate your research?

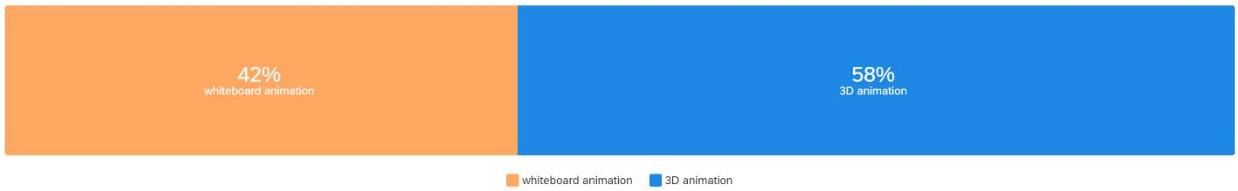
Q50 - How clear was the information presented in this animation? Page Options



#	Field		Choice Count
5	Slightly unclear	12.50%	3
3	Slightly clear	33.33%	8
4	Neither clear nor unclear	8.33%	2
6	Moderately unclear	0.00%	0
2	Moderately clear	37.50%	9
7	Extremely unclear	0.00%	0
1	Extremely clear	8.33%	2
			24

Figure 74. Low spatial reasoning participant response to: How clear was the information presented in this [whiteboard] animation?

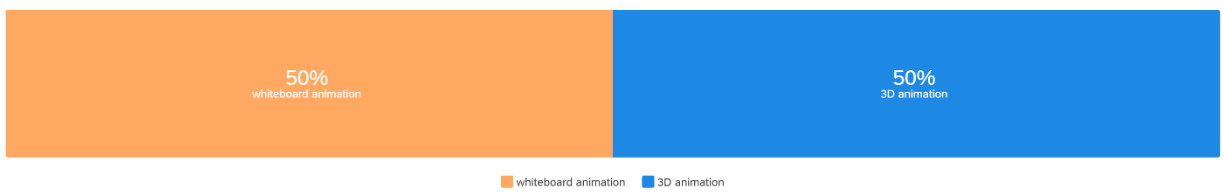
Q54 - I prefer to use the ________ as a learning tool. Page Options



#	Field	Choice Count	
1	whiteboard animation	41.67%	10
2	3D animation	58.33%	14
			24

Figure 75. Low spatial reasoning participant response to: I prefer to use ___ as a learning tool.

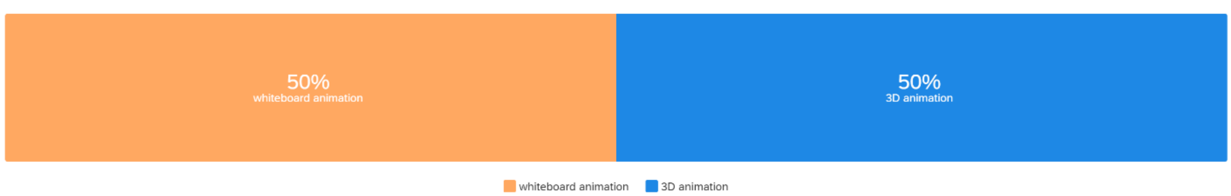
Q55 - I enjoyed the __________ most. Page Options



#	Field	Choice Count	
1	whiteboard animation	50.00%	12
2	3D animation	50.00%	12
			24

Figure 76. Low spatial reasoning participant response to: I enjoyed the ___ most

Q56 - The _________ is more successful at communicating complex information. Page Options

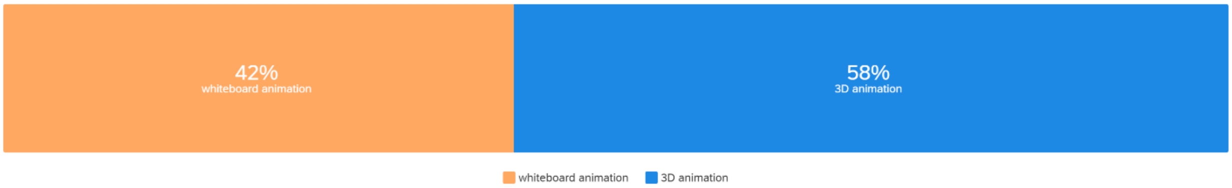


#	Field	Choice Count	
1	whiteboard animation	50.00%	12
2	3D animation	50.00%	12
			24

Figure 77. Low spatial reasoning participant response to: The ___ is more successful at communicating complex information

Q57 - I am more likely to recommend the __________ to peers.

Page Options

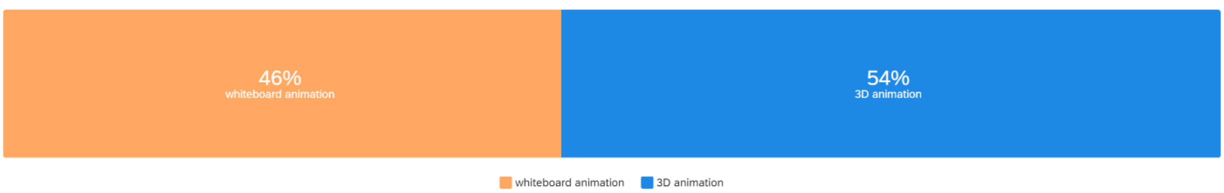


#	Field	Choice Count	
1	whiteboard animation	41.67%	10
2	3D animation	58.33%	14
			24

Figure 78. Low spatial reasoning participant response to: I am more likely to recommend ___ to peers.

Q58 - The _____ was easiest to understand.

Page Options



#	Field	Choice Count	
1	whiteboard animation	45.83%	11
2	3D animation	54.17%	13
			24

Figure 79. Low spatial reasoning participant response to: The ___ was easiest to understand.

Q51 - Rate your level of knowledge with bacterial signaling.

Page Options

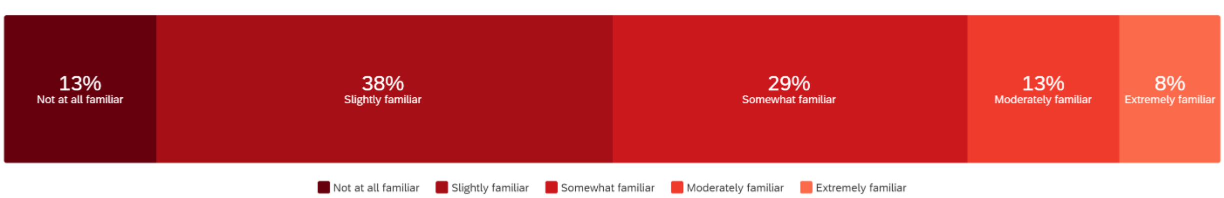
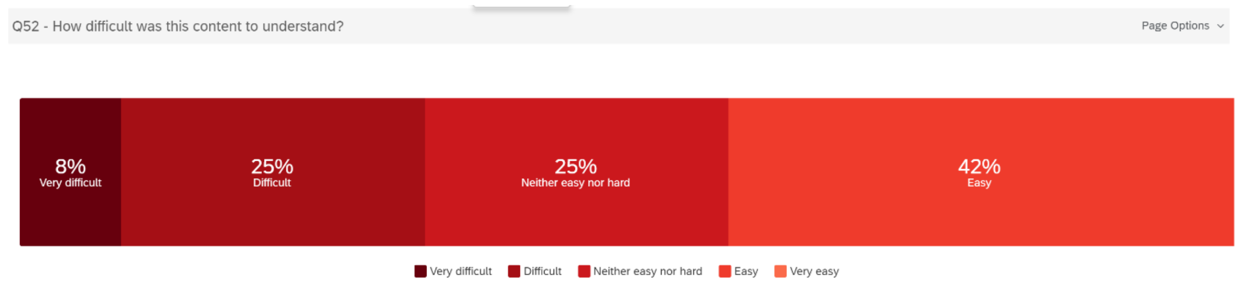


Figure 80. Low spatial reasoning participant response to: Rate your level of knowledge with bacterial signaling



#	Field	Minimum	Maximum	Mean	Std Deviation	Variance	Count
1	How difficult was this content to understand?	1.00	4.00	3.00	1.00	1.00	24

Figure 81. Low spatial reasoning participant response to: How difficult was the content to understand?

Figure 82. Low spatial reasoning participant response to: How much effort was required to comprehend the content?

VI. DISCUSSION

A. Review of major points

The project resulted in a 3D computer-based animation and a 2D whiteboard animation showing how bacterial electrochemical communication in biofilms works. The primary objectives of the project are to evaluate the 3D animation and the whiteboard animation for engagement, preference, and knowledge gained on a sample population of students primarily in the fields of science and/or medicine. Calculating the Pearson's correlation coefficients between knowledge transfer and engageability, spatial reasoning, and self-reported cognitive load would allow for the assessment of how these variables are related and whether these relationships are statistically significant. Next, a breakdown of the results by viewer preference and level of knowledge of bacterial signaling would provide key insights as to how participant responses aggregated across these variables.

It was expected that there would be strong positive correlations between knowledge transfer and engageability, knowledge transfer and spatial reasoning, and knowledge transfer and self-reported cognitive load. However, not all these calculations returned what was expected. Calculations of Pearson's correlation coefficient on knowledge transfer and spatial reasoning scores revealed a statistically significant moderate positive correlation, indicating that higher knowledge transfer scores tend to go with higher spatial reasoning scores. Although Pearson's correlation coefficient revealed a statistically significant positive correlation between knowledge transfer and engagement scores, the correlation is weak. The weak positive correlation found between knowledge transfer and self-reported cognitive load did not provide a statistically significant p-value.

Of the two-tailed t-tests of two independent means, the only statistically significant result was that viewers who preferred the whiteboard animation reported significantly lower levels of cognitive load than the viewers who preferred the 3D animation. This exciting result implies that the intent of creating a whiteboard animation which conveys the same information as the 3D animation without risking cognitive overload was a success.

When comparing participants with low levels of prior knowledge versus participants with high levels of prior knowledge, it was found that the former overwhelming preferred the whiteboard animation, which was expected, and the latter preferred the 3D animation, which was not expected. Surprisingly, low spatial reasoning was not as strong an indicator for whiteboard animation preference as low prior knowledge. In fact, the low spatial reasoning population seemed more or less evenly split on preference, while veering slightly more towards the 3D animation.

B. Limitations

This study is limited by four primary factors. First, time: a significant amount of time was required to construct the stimuli. Time limitations also affected sample size, since only a small amount of time could be spent recruiting volunteers and testing. Second, my own technical expertise, since I am just now learning 3D animation and teaching myself whiteboard animation concurrently. My own lack of experience compounds the time constraints I face, especially with the addition of the 3DSMax TyFlow plugin. Third, the money required for testing, including purchasing equipment and payment required for using Mechanical Turk. Fortunately, due to the hardware and software provided by the school, and because of the relatively low costs of whiteboard animation, financial limitations were minimal. The final limitation is the replicability of my research, since others may not have the capability for whiteboard animation. Many whiteboard animations rely on unique stylistic drawings, so different artists would develop different characteristics of whiteboard animation based on their own personal illustration approaches.

C. Implications for profession

This project's results may inform best practices for whiteboard animation relying on the conceptual frameworks of human cognition and multimedia learning. This project may also lead to further applications of whiteboard animation in biocommunication between health professionals and scientists. Whiteboard animation provides a more accessible threshold animation in that it can be less demanding on time, resources, money, and technical expertise, and it can potentially avoid cognitive overload, providing a highly efficient mechanism of biocommunication.

D. Future applications

This research can be applied to other inherently complex and interdisciplinary topics which require communication between scientists and health professionals. For scientists who struggle communicating their research with other educated audiences, whiteboard animation may provide an efficient, accessible, and aesthetically appealing mode of communication. Additionally, best practices for whiteboard-style animation have yet to be defined and the medium lacks sufficient academic research to support its popularity.

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APPENDIX A

Class Announcement

[The investigator will visit the lecture sections during the first week of testing and make this announcement during the spring semester of 2020 to the class of: ANAT 441, GEMS 502]

Good morning and thank you, Professor _______, for allowing me to speak with your class today. My name is Sarah McGuinness and I am a Master's student in the Biomedical Visualization program with the College of Applied Health Sciences. I create visual media such as illustrations and animations to teach science.

Please raise your hand if you have ever watched whiteboard-style videos, like Khan Academy or RSA Animate, to study for a class. [pause]

Please raise your hand if you have ever watched whiteboard-style videos just for fun. [pause]

Thank you.

As part of my research project, I created animations about bacterial signaling for higher education audiences and I have developed a research project here at UIC that is investigating preference between 3D and whiteboard animation visual style and knowledge gained. You are invited to participate in an anonymous digital survey that will test these animations. You are being asked because of your level of prior knowledge with bacterial signaling.

You may be wondering why this is even important. What is the point of testing this? The issue that led me to this research is the difficulty of clear and efficient communication within and between the scientific community and health professionals. Many public health issues involve the efforts of individuals from a wide range of disciplines, so effective communication is key. It is my hope to continue to work to help develop effective communication and learning resources for people who work in highly specified fields.

An announcement will be posted on the Blackboard site with a link. By clicking on that link you may fill out a consent form to participate in this research. You will be asked to answer some questions before and after watching an animation. You will then be asked to watch an alternate style animation and answer survey questions. Please feel free to contact me with any questions. Thank you in advance for your participation.

APPENDIX B

Class Handout

[The investigator will visit the lecture sections during the first week of testing and will hand out this information during the spring semester of 2020 to the class of: ANAT 441 and MIM 560]

Front:

[image]

Back:
Hi! As part of my research project, I created animations about bacterial signaling for higher education audiences and I have developed a research project here at UIC that is investigating preference between 3D and whiteboard animation visual style and knowledge gained. You are invited to participate in an anonymous digital survey that will test these animations. For more information, contact me at smcgui5@uic.edu.
Sincerely, Sarah McGuinness, Department of Biomedical Visualization

APPENDIX C

Class Email

Dear Students,

You are invited to participate in an anonymous digital survey that will test animations about bacterial signaling for higher education audiences. You will be able to provide valuable feedback about the design choices and knowledge gained in creating an animation for audiences in higher education.

By clicking on the link provided below you may fill out a consent form to participate in this research. You will be asked to answer some questions before and after watching an animation. You will then be asked to watch an alternate style animation and answer survey questions.

If you are interested in participating in this research, please click the following link to participate: [Qualtrics link]

Attached to this email is the informed consent form. Please feel free to contact me with any questions.

Thank you in advance for your participation!

Sincerely,
Sarah McGuinness, Department of Biomedical Visualization
Smcgui5@uic.edu

APPENDIX D

MTurk Task Explanation

[This will be the task explanation after workers have accepted the HIT (Human Intelligence Task)]

You are invited to participate in an anonymous digital survey that will test animations about bacterial signaling for higher education audiences. You are being asked because the animation was created for an audience in the biomedical sciences and health professions and we are interested in getting a large and varied population response. You will provide valuable feedback about the design choices and knowledge gained in creating an animation for the general public.

By clicking on the link provided below you may fill out a consent form to participate in this research. You will be asked to answer some questions before and after watching an animation. You will then be asked to watch an alternate style animation and answer survey questions.

If you are interested in participating in this research, please click the following link to participate: [Qualtrics link]

Please feel free to contact me with any questions. Thank you in advance for your participation!

Sincerely,
Sarah McGuinness, Department of Biomedical Visualization
smcgui5@uic.edu

APPENDIX E

Snowballing Request

[After completing the study, participants will be taken to a “Thank You” message with the text below. Participants can leave the web page at any point and no further action will be needed on their part to complete the study.]

Thank you for participating in this research. We hope to use the information gathered from this research to inform creation of more educational animations for the general public. The more people who participate, the better we can make these materials. We hope you would be willing to pass along information about this study to friends and/or family members who may also be interested in learning about this research study. You are under no obligation to share this information.
If you would like to share this study please share the following link: [Qualtrics link]
If you have any questions, please contact either:

Primary Investigator:
Sarah McGuinness
Smcgui5@uic.edu

Faculty Sponsor:
Christine Young, MA, CMI, FAMI
cdy@uic.edu

Sincerely,
Sarah McGuinness

APPENDIX F

Knowledge Transfer (Pre and Post) Test

1) A community of bacteria is called a(n) _________.
   a) microorganism
   b) collective
   c) **biofilm**
   d)
2) The most well-known form of bacterial communication is _____
   a) **quorum sensing**
   b) electrochemical signaling
   c) ion signaling
   d) signal transduction
3) Electrochemical signaling in biofilms is enabled by___________
   a) symporters
   b) neurons
   c) the cell membrane
   d) **ion channels**
4) Q: Neuronal signals travel _____ bacterial electrochemical signaling.
   a) **Faster than**
   b) Slower than
   c) At the same speed as
   d) None of the above: relative speeds vary widely.
5) ________ cells protect the biofilm while ______ cells are responsible for maintaining the biofilm following external attack
   a) Interior, peripheral
   b) Target, attachment
   c) **Peripheral**, **interior**
   d) Peripheral, attachment
6) _________ is the first step in biofilm formation.
   a) Attachment
   b) Dispersal
   c) Monolayer Formation
   d) Signaling
7) Production of a(n) _______ holds individual cells in the biofilm together.
   a) Lipopolysaccharide membrane
   b) **Extracellular matrix**
   c) Electrochemical signal
   d) Gap junction
8) Bacterial electrochemical signaling was discovered in _______.
   a) *Pseudomonas aeruginosa*
   b) *Escherichia coli*
   c) *Staphylococcus aureus*
   d) ***Bacillus subtilis***

9) _____________ can describe the movement of signals through a biofilm.
   a) Discretization
   b) **Percolation theory**
   c) Signal transduction
   d) Extracellular flow
10) _______% of chronic infections are biofilm-associated.
   a) 90%
   b) 60%
   c) **80%**
   d) 30%
11) Electrochemical signaling can coordinate bacteria over long distances.
   a) True
   b) False
12) Neuronal signals travel along the length of the cell's axon, whereas bacterial electrochemical signaling travels _______.
   a) **from cell to cell**
   b) from ion channel to ion channel
   c) along the cell's body
   d) along the cellular membrane
13) Select the correct order of steps in biofilm formation:
   a) Secretion, attachment, maturation, monolayer formation, migration
   b) Attachment, secretion, monolayer formation, migration, maturation
   c) Attachment, maturation, monolayer formation, migration, secretion
   d) Attachment, monolayer formation, migration, secretion, maturation

APPENDIX G

Demographic Questions

Please fill out the demographic questions below as accurately as possible. This survey will remain anonymous. In order to move on to the next window, you must answer all the questions below.

1) Select your gender:
    a) Male
    b) Female
    c) Nonbinary
    d) Other: _________
2) Select your age range:
    a) 18-24
    b) 25-34
    c) 45-54
    d) 55-64
    e) 65-74
    f) 75 or older
3) Select your region of residence:
    a) North America
    b) South America
    c) Central America
    d) Europe
    e) Asia
    f) Africa
4) Select your education level:
    a) High school
    b) Bachelor's degree
    c) Master's Degree
    d) Doctorate degree
5) Which of these college-level classes have you previously taken? [checkbox entry]
    a) Biology
    b) Chemistry
    c) Microbiology
    d) Neuroscience
    e) Biochemistry
    f) Molecular Biology
    g) Bacterial Genetics
6) In which field do you currently work or study?
    a) Medicine
    b) Pharmacy
    c) Nursing
    d) Public Health
    e) Biological Sciences
    f) Chemical Sciences
    g) Social Sciences

h) Biomedical Visualization
i) Other: ___________

7) Rate your level of prior knowledge with bacterial signaling.
   a) Not at all familiar
   b) Slightly familiar
   c) Somewhat familiar
   d) Moderately familiar
   e) Extremely familiar

APPENDIX H

Modified Engageability Framework

Answer the questions below on a seven point scale: How accurately does this statement describe you?

1. Very untrue of me
2. Untrue of me
3. Somewhat untrue of me
4. Neutral
5. Somewhat true of me
6. True of me
7. Very true of me

1) I feel comfortable looking at most types of animations.
2) I like to know about the story portrayed in an animation.
3) I like to know about the materials and techniques used by the animators.
4) I enjoy talking with others about the animation we are looking at.
5) I am emotionally affected by animation.
6) I like to be provided with text to help me understand what the animation is about.
7) I like to look at animation.
8) I am comfortable explaining an animation to a friend.
9) I find some animations difficult to understand.

APPENDIX I

Post-Stimulus Survey for Whiteboard Animation

1. How useful is this animation for understanding bacterial electrochemical signaling?
    1.1. Extremely useless
    1.2. Somewhat useless
    1.3. Slightly useless
    1.4. Neither useful nor useless
    1.5. Slightly useful
    1.6. Somewhat useful
    1.7. Extremely useful
2. How strongly do you agree or disagree with this statement: I enjoyed the 2D quality of the whiteboard animation.
    2.1. Strongly disagree
    2.2. Disagree
    2.3. Somewhat disagree
    2.4. Neither agree or disagree
    2.5. Somewhat agree
    2.6. Agree
    2.7. Strongly agree
3. How strongly do you agree or disagree with this statement: I enjoyed the simplicity of the whiteboard animation.
    3.1. Strongly disagree
    3.2. Disagree
    3.3. Somewhat disagree
    3.4. Neither agree or disagree
    3.5. Somewhat agree
    3.6. Agree
    3.7. Strongly agree
4. How likely are you to use whiteboard animation to communicate your research?
    4.1. Extremely unlikely
    4.2. Somewhat likely
    4.3. Unlikely
    4.4. Neither likely nor unlikely
    4.5. Likely
    4.6. Somewhat likely
    4.7. Extremely likely
5. How clear was the information presented in this animation?
    5.1. Extremely unclear
    5.2. Slightly unclear
    5.3. Neither clear nor unclear
    5.4. Slightly clear
    5.5. Somewhat clear
    5.6. Extremely clear

APPENDIX J

Post-Stimulus Survey for 3D Animation

1. How useful is this animation for understanding bacterial electrochemical signaling?
    1.1. Extremely useless
    1.2. Somewhat useless
    1.3. Slightly useless
    1.4. Neither useful nor useless
    1.5. Slightly useful
    1.6. Somewhat useful
    1.7. Extremely useful
2. How strongly do you agree or disagree with this statement: I enjoyed the 3D quality of the animation.
    2.1. Strongly disagree
    2.2. Disagree
    2.3. Somewhat disagree
    2.4. Neither agree or disagree
    2.5. Somewhat agree
    2.6. Agree
    2.7. Strongly agree
3. How strongly do you agree or disagree with this statement: I enjoyed the complexity of the 3D animation.
    3.1. Strongly disagree
    3.2. Disagree
    3.3. Somewhat disagree
    3.4. Neither agree or disagree
    3.5. Somewhat agree
    3.6. Agree
    3.7. Strongly agree
4. How likely are you to use 3D animation to communicate your research?
    4.1. Extremely unlikely
    4.2. Somewhat likely
    4.3. Unlikely
    4.4. Neither likely nor unlikely
    4.5. Likely
    4.6. Somewhat likely
    4.7. Extremely likely
5. How clear was the information presented in this animation?
    5.1. Extremely unclear
    5.2. Slightly unclear
    5.3. Neither clear nor unclear
    5.4. Slightly clear
    5.5. Somewhat clear
    5.6. Extremely clear

APPENDIX K

Survey Comparing Both Stimuli

1. I prefer to use the whiteboard animation of the 3D animation as a learning tool.
   1.1. Strongly disagree
   1.2. Disagree
   1.3. Somewhat disagree
   1.4. Neither agree or disagree
   1.5. Somewhat agree
   1.6. Agree
   1.7. Strongly agree
2. I enjoyed the whiteboard animation more than the 3D animation.
   2.1. Strongly disagree
   2.2. Disagree
   2.3. Somewhat disagree
   2.4. Neither agree or disagree
   2.5. Somewhat agree
   2.6. Agree
   2.7. Strongly agree
3. The whiteboard animation is more successful than the 3D animation at communicating complex information.
   3.1. Strongly disagree
   3.2. Disagree
   3.3. Somewhat disagree
   3.4. Neither agree or disagree
   3.5. Somewhat agree
   3.6. Agree
   3.7. Strongly agree
4. I am more likely to recommend the whiteboard animation than the 3D animation to peers.
   4.1. Extremely unlikely
   4.2. Somewhat likely
   4.3. Unlikely
   4.4. Neither likely nor unlikely
   4.5. Likely
   4.6. Somewhat likely
   4.7. Extremely likely
5. The whiteboard animation was clearer than the 3D animation.
   5.1. Strongly disagree
   5.2. Disagree
   5.3. Somewhat disagree
   5.4. Neither agree or disagree
   5.5. Somewhat agree
   5.6. Agree
   5.7. Strongly agree

APPENDIX L

Perceived Mental Difficulty

1. Rate your level of knowledge with bacterial signaling.
    1.1. Not at all familiar
    1.2. Slightly familiar
    1.3. Somewhat familiar
    1.4. Moderately familiar
    1.5. Extremely familiar
2. How difficult was this content to understand?
    2.1. Very difficult
    2.2. Difficult
    2.3. Neither easy nor hard
    2.4. Easy
    2.5. Very easy
3. How much effort was required to comprehend the content?
    3.1. Very high effort
    3.2. High effort
    3.3. Some effort
    3.4. Little effort
    3.5. Very little effort

APPENDIX M

Script

TITLE	The Bacterial Connection
VERSION	Final
DATE	02/12/2020
EST. RUNTIME	3 minutes
BY	Sarah McGuinness
FOR	Dr. Prindle
AUDIENCE	Graduate level
PURPOSE	Explain how bacterial electrochemical signaling works and why it is important.
WORDS	348

<u>Intro Scene</u>

- Biofilms are everywhere. Defined as a collective of microorganisms that adhere to a surface, they impact our lives in a myriad of ways.
- There is a medical need to understand their communication and growth, because 80% of chronic and hard to treat infections involve bacterial biofilms.
- Recent lab investigations have uncovered a novel method of bacterial communication in isolated homogenous biofilms of the Gram-positive bacteria *Bacillus subtilis*.

<u>Scene 1</u>

- Biofilms form through a tightly regulated process.
- First, a single bacterial cell attaches to a wet surface.
- As bacteria begin dividing, a monolayer forms.
- Migration forms more layers.
- After migration, the bacteria secrete a glue-like extracellular matrix of glycoproteins.
- Finally, upon maturation, the biofilm differentiates according to location within the biofilm community.

<u>Scene 2</u>

- The most well-known form of biofilm communication is quorum sensing.
  - This extracellular communication depends on the release of small chemical signals which coordinate bacterial behavior.
- Scientific research cell to cell bacterial signaling dependent on potassium ions and their ion channels to communicate over long distances.
  - Signals travel cell to cell, like oil through cracks in the earth, via percolation.
- This process is very similar to the signals which propagate along neurons, yet more slowly.
  - Neuron signals travel along the length of the cell's axon.
- This new form of bacterial communication travels from cell to cell over a long distance.
  - But why does this happen?

<u>Scene 3</u>

- Biofilms have two distinct groups of cells: peripheral cells and interior cells.

- Peripheral cells have more access to nutrients and serve to protect the biofilm from external attack.
- Interior cells have limited access to nutrients but can survive and maintain biofilm growth if the peripheral cells are destroyed.
- Also, cellular location within the biofilm creates gradients of oxygen and nutrients, establishing a need for metabolic coordination and necessitating more sophisticated methods of communication.

Concluding Scene

- Over 17 million cases of biofilm associated infections occur every year in the US, resulting in over 500,000 deaths and an annual cost of $94 billion. Further understanding of bacterial communication may hold the keys to both preventing and fighting these complex infections.

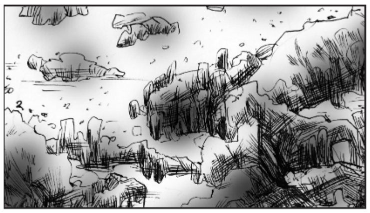
APPENDIX N

Storyboard for 3D Animation

TITLE: The Bacterial Connection

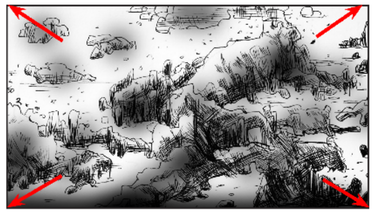
PAGE: 2

2-1



AUDIO: ...of microorganisms that adhere ...
ANIM: Camera pans right and zooms out.

2-2



AUDIO: ... to a surface, ...

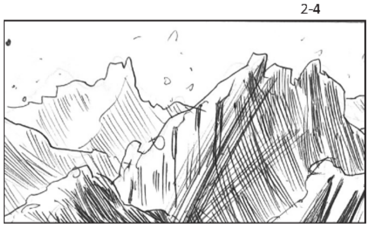
ANIM: Camera pans right and zooms out.

2-3

AUDIO: ... they impact our ...

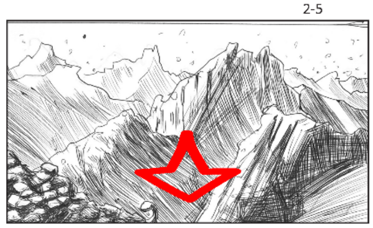
ANIM: Camera pans right and zooms out.

2-4



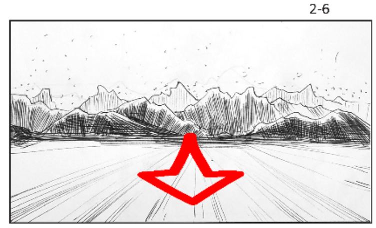
AUDIO: ... lives...
ANIM: Camera pans right and zooms out.

2-5



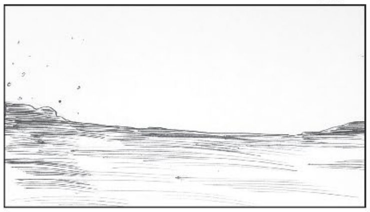
AUDIO: ... in a myriad ...
ANIM: Camera trucks backwards.

2-6



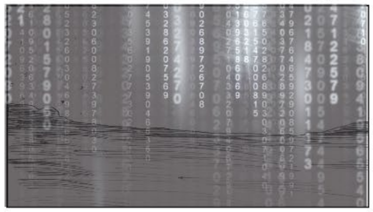
AUDIO: ... of ways.
ANIM: Camera trucks backwards.

3-1



AUDIO: There is a medical need to understand bacterial biofilm communication ...
ANIM: Camera trucks backwards.

3-2



AUDIO: ... because. ..
ANIM: Numbers fall down over the screen. Camera zooms in.

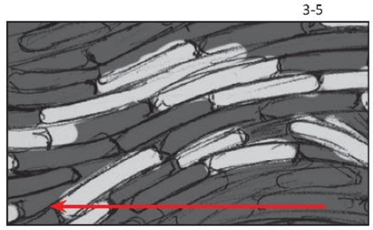
3-3

AUDIO: ... 80% of chronic and hard to treat infections involve bacterial biofilms.
ANIM: "80%" appears onscreen as camera zooms in.

3-4

AUDIO: Recent lab investigations have uncovered a novel method ...
ANIM: Camera pans left to follow percolating signals.

3-5

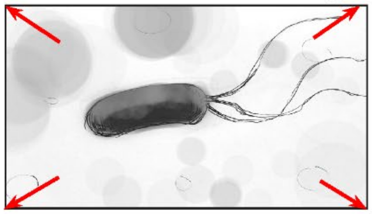


AUDIO: ... of bacterial communication ...
ANIM: Camera pans left to follow percolating signals.

3-6

AUDIO: ... in isolated homogenous biofilms of ...
ANIM: Zoom in as all but one cell become unfocused.

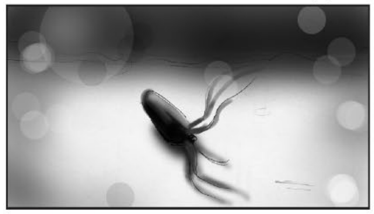
4-1



AUDIO: ...the Gram-negative bacteria Bacillus subtilis.

ANIM:Previous scene dissolves into scene of single cell swimming using flagella. Camera pans left to follow bacteria and zooms out.

4-2

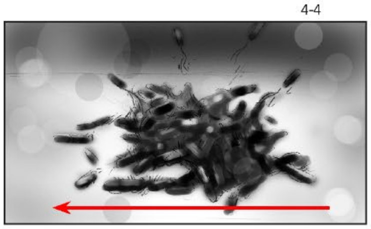


AUDIO: Biofilms ...
ANIM:

4-3

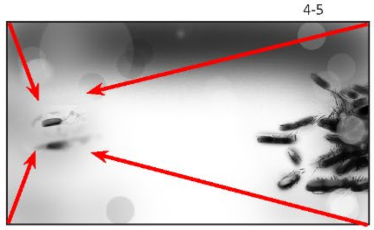
AUDIO: ... form through ...
ANIM:

4-4



AUDIO: ... a tightly regulated process.
ANIM: Camera pans left.

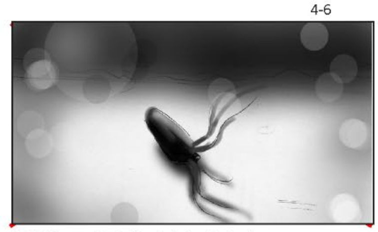
4-5



AUDIO: First ...

ANIM: Camera pans left as another single bacteria approaches the surface.

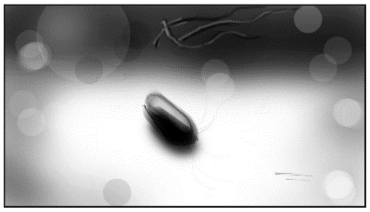
4-6



AUDIO: ... a single bacterial cell attaches ...

ANIM: Camera zooms in as bacteria delicately lands on the surface.

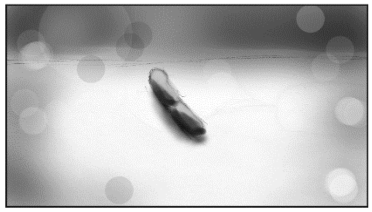
5-1



AUDIO: to a wet surface.

ANIM: The bacterial cell's flagella detaches.

5-2



AUDIO: As bacteria begin dividing ...

ANIM: Cell divides into two.

5-3

AUDIO: ... a monolayer ...

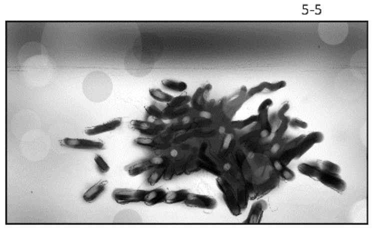
ANIM: Cells continue to divide.

5-4

AUDIO: ... forms.

ANIM: Cells continue to divide.

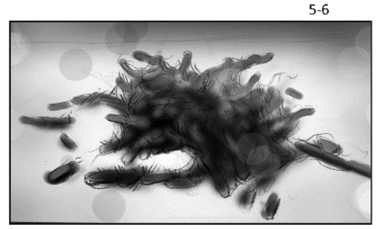
5-5



AUDIO: Migration forms more layers.

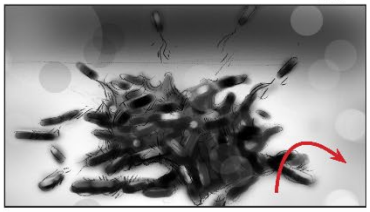
ANIM: Colony continues to divide, and cell spread to occupy the spaces above the initial layer.

5-6



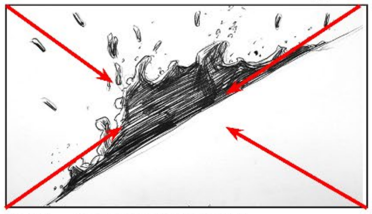
AUDIO: After migration, the bacteria secrete a glue-like extracellular matrix of glycoproteins.
ANIM: The colony continues expanding. A fibrous, substance is secreted and binds the collective together.

6-1



AUDIO: Finally, upon maturation, the cells differentiate according to ...
ANIM: Individual cells of the coony turn slightly different colors to represent differentiation upon maturation.
Individual cells develop flagella and disperse from the biofilm.

6-2



AUDIO: ... location within the biofilm ...
ANIM: Camera tilts and zooms out to see sagittal section of biofilm.

6-3

AUDIO: ...community.

ANIM: Camera zooms in rapidly. Transition to next scene

6-4

AUDIO: The most well known ..

ANIM: Camera slowly zooms out and rotates CCW.

6-5

AUDIO: ... form of biofilm communication is ...

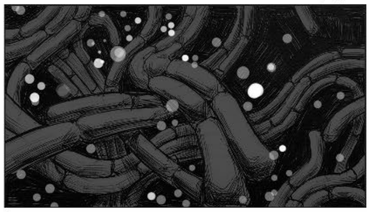
ANIM: Camera slowly zooms out and rotates CCW.

6-6

AUDIO: ... quorum sensing.

ANIM: Camera slowly zooms out and rotates CCW.

7-1



AUDIO: This extracellular communication depends on the release of small chemical signals to coordinate bacterial behavior.
ANIM: Chemical signals are released from the bacteria.

7-2

AUDIO: Scientific research ...

ANIM: Truck backwards. Chains of bacteria come into view and focus.

7-3

AUDIO: ... has investigated ...

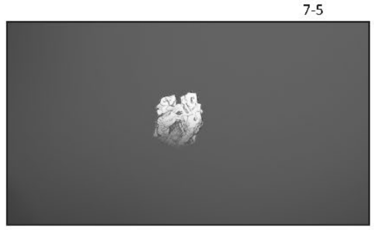
ANIM: Truck backwards. Chains of bacteria come into view and focus.

7-4

AUDIO: ... cell to cell ....

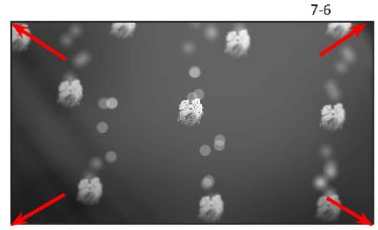
ANIM: Camera zooms in onto surface of bacterial cell.

7-5



AUDIO: ... bacterial signaling dependent on potassium ions and their ion channels to communicate over long distances.
ANIM:
Camera zooms in onto ion channel.

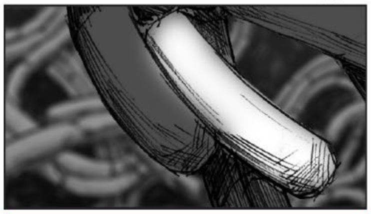
7-6



AUDIO: Signals travel from cell to cell, ...

ANIM: Potassium ions exit the ion channel as the signal begins.

8-1



AUDIO: ... like oil ...

ANIM: Camera zooms out as signal initiates. The first cell glows.

8-2

AUDIO: ... through cracks ...

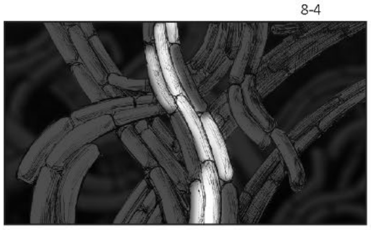
ANIM: Camera zooms out as signal propagates from cell to cell.

8-3

AUDIO: ... in ...

ANIM: Camera zooms out and trucks to follow signal propagation.

8-4



AUDIO: ...the ...

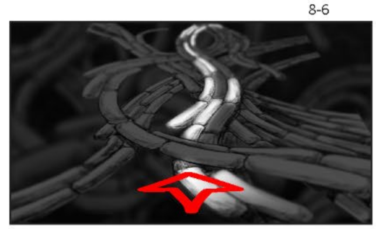
ANIM: Camera trucks to follow signal. Camera orbits upwards.

8-5

AUDIO: ... earth ...

ANIM: Camera trucks forwards to follow signal.

8-6



AUDIO: ... via ...

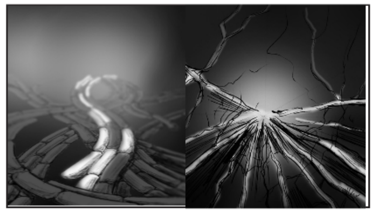
ANIM: Camera trucks forwards to follow signal.

9-1

AUDIO: ... percolation theory.

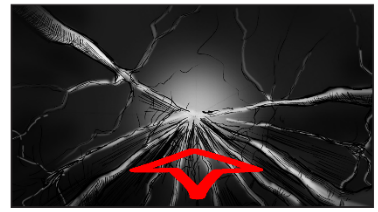
ANIM: Camera trucks forwards to follow signal.

9-2



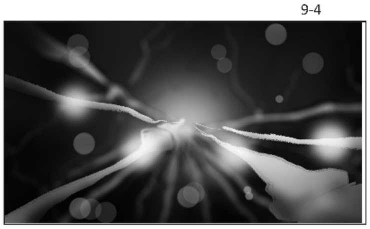
AUDIO: This process is very similar to the signals which propagate along neurons, yet more slowly.
ANIM: Neuron scene slides in from the right: split screen showing camera trucking through bacterial signaling scene and neuron signaling scene.

9-3



AUDIO: Neuron signals travel ...
ANIM: Camera trucks forwards.

9-4



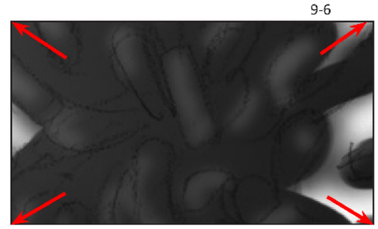
AUDIO: ... along the length of the cell's axon.

ANIM: Camera trucks forwards, passing by signals propagating. Scene darkens to transition.

9-5

AUDIO: This new form of bacterial communication travels from cell to cell over a long distance.
ANIM: Cut to bacterial signaling scene. Scene darkens to transition.

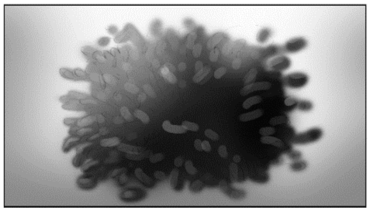
9-6



AUDIO: But why does this happen?

ANIM: Cut to close-up of small biofilm in sterile environment. Camera zooms out.

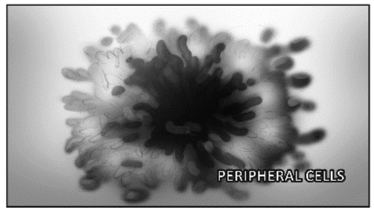
10-1



AUDIO: Biofilms have two distinct groups of cells: ...

ANIM: Camera zooms out onto scene of fully grown biofilm.

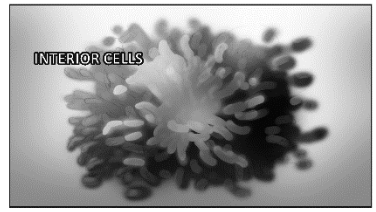
10-2



AUDIO: ... peripheral cells ...

ANIM: Peripheral cells glow.

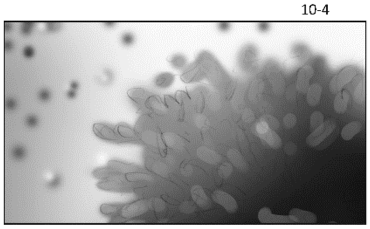
10-3



AUDIO: and interior cells.

ANIM:
Interior cells glow.

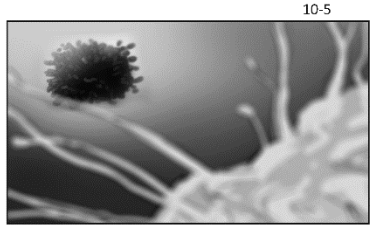
10-4



AUDIO: Peripheral cells have more access to nutrients ...

ANIM: Cut to close-up of biofilm with glowing particles approaching, representing nutrients.

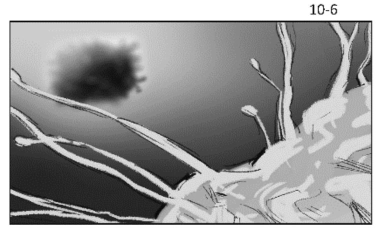
10-5



AUDIO: .. and serve to protect ...

ANIM: Cut to view of biofilm from afar. Phagocyte appears from behind camera menacingly.

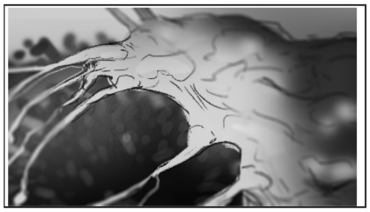
10-6



AUDIO: ... the biofilm from external attack.

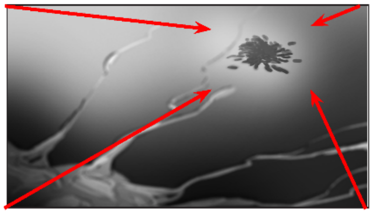
ANIM: Rack focus to focus on phagocyte as it approaches.

11-1



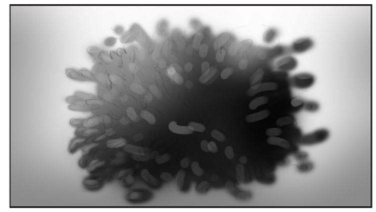
AUDIO: Interior cells have limited access to nutrients, but can survive and maintain biofilm growth ...
ANIM: Camera cut to phagocyte devouring biofilm.

11-2



AUDIO: ... if the peripheral cells are destroyed.
ANIM: Phagocyte crawls over biofilm and away, leaving destruction in its wake. Camera cuts to view of interior cells in the distance.

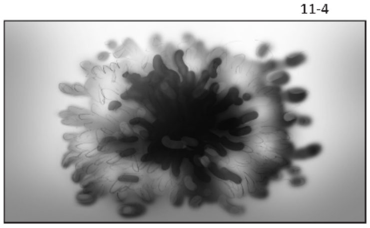
11-3



AUDIO: Cellular location ..

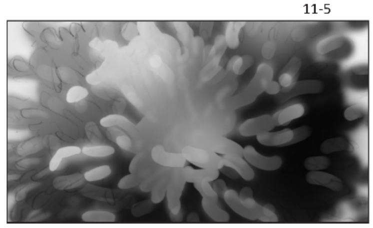
ANIM: Zoom in onto interior cells, which are already growing into a restored biofilm.

11-4



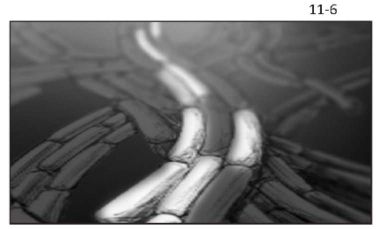
AUDIO: ... within the biofilm creates gradients of oxygen and nutrients ...
ANIM: Outer and inner cells glow alternatively. Growth of the biofilm is scene to oscillate.

11-5



AUDIO: ... establishing a need for metabolic codependency ...
ANIM: Glow/growth continues to oscillate. Camera zooms in rapidly to transition to next scene.

11-6

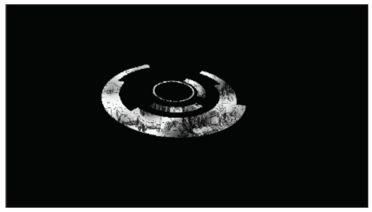


AUDIO: ... necessitating more sophisticated methods of communication.
ANIM: Transition to scene of bacterial signaling. Cut to next scene.

12-1

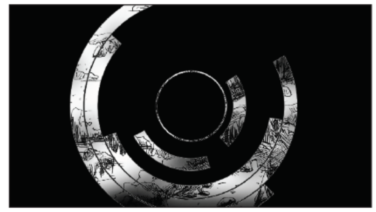
AUDIO: Over 17 million cases of biofilm associated infections occur every year in the US ...
ANIM: Gradual zoom as white dots appear on black background, gradually forming shape of the US map. Camera moves forward through map.

12-2



AUDIO: ... resulting in over 500,000 deaths and an annual cost of $94 billion.
ANIM: Animated radial bar chart moves in 3D towards viewer.

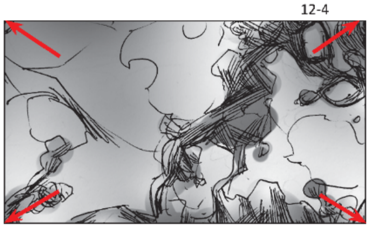
12-3



AUDIO: Further understanding...

ANIM: Camera trucks forwards through radial bar chart.

12-4

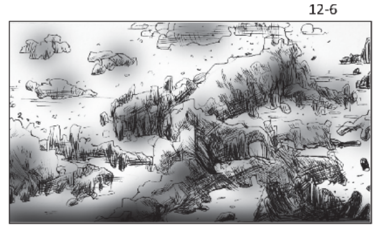


AUDIO: ... of bacterial communication and growth may hold the keys ...
ANIM: Particle flow data operators used to form surface architecture which resembles heterogenous biofilm growth. Camera zooms out.

12-5

AUDIO: ...to both preventing and fighting these complex infections.
ANIM: Camera zooms out to reveal that the surface architecture has formed in the shape of a key.

12-6



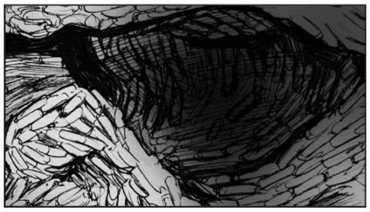
AUDIO:

ANIM: Credits play.

1-1

AUDIO: Biofilms ...
ANIM: Camera slowly zooms out and pans right from close-up of bacterial biofilm.

1-2



AUDIO: ... are ...

ANIM: Camera slowly zooms out and pans right from close-up of bacterial biofilm.

1-3

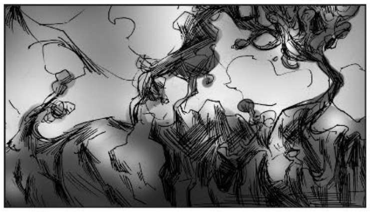
AUDIO: everywhere.
ANIM: Camera zooms out.

1-4

AUDIO: Defined ...

ANIM: Camera zooms out onto architecture of biofilm and pans right slowly.

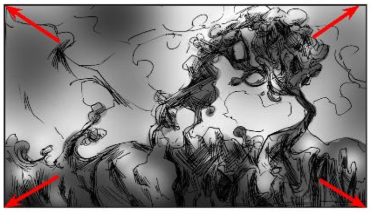
1-5



AUDIO: ...as...

ANIM: Camera slowly zooms out and pans right.

1-6



AUDIO: ...a collective ...
ANIM: Camera zooms out.

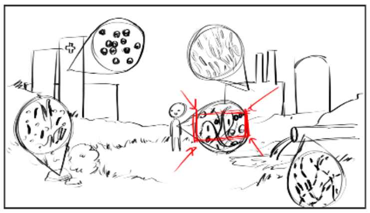
APPENDIX O

Storyboard for Whiteboard Animation

TITLE: The Bacterial Connection

PAGE: 1

1-1

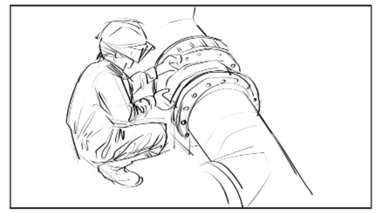


AUDIO: Biofilms are everywhere. Defined as a collective of microorganisms that adhere to a surface,....
ANIM: Hand draws a person standing in the middle of a scene that represents medical, industrial, and natural processes. Insets showing various biofilms are drawn.

1-2

AUDIO: ... they impact our lives in a myriad of ways.
ANIM: Camera pans right and hand draws a scene of a patient in a hopsital.

1-3

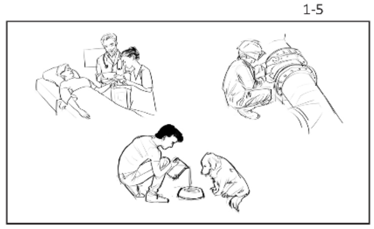


AUDIO: ... There is a medical need ...
ANIM: Camera pans right and hand draws a scene of an engineer working.

1-4

AUDIO: ... need to understand ...
ANIM: Camera pans right and hand draws a scene of an individual filling his dog's water bowl.

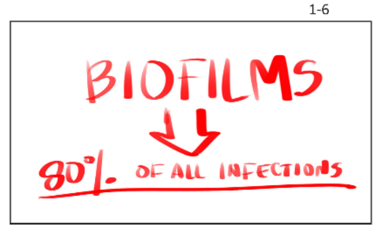
1-5



AUDIO: ... their communication and growth, ...

ANIM: Camera zooms out to show all three scenes together.

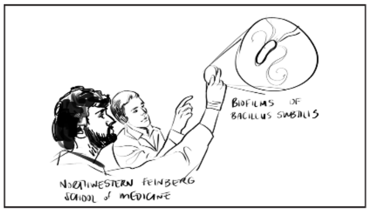
1-6



AUDIO: ...because 80% of chronic and hard to treat infections involve bacterial biofilms.
ANIM: Camera pans right as text is written onscreen.

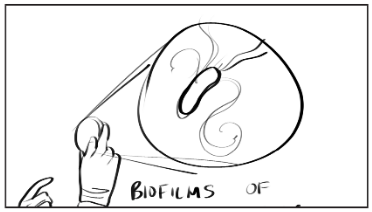
TITLE: The Bacterial Connection PAGE: 2

2-1



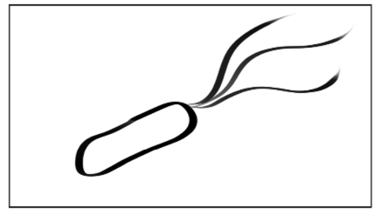
AUDIO: Recent lab investigations have uncovered a novel method of bacterial communication in isolated homogenous biofilms of the Gram-positive bacteria Bacillus subtilis.
ANIM: Camera pans right and scene of two scientists in a lab holding up an agar plate is drawn.

2-2



AUDIO: Biofilms form through a tightly regulated process.
ANIM: Camera zooms in onto inset of plate to show flagelatted bacteria.

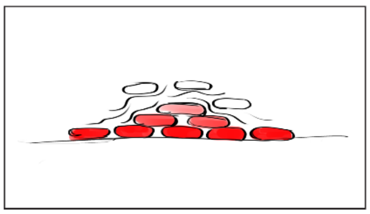
2-3



AUDIO: First, a single bacterial cell attaches to a wet surface.

ANIM: Lines surrounding bacteria are erased.

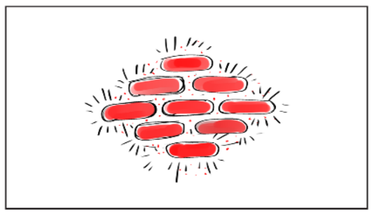
2-4



AUDIO: First, a single bacterial cell attaches to a wet surface. As bacteria begin dividing, a monolayer forms. Migration forms more layers. After migration, the bacteria secrete a glue-like extracellular matrix of glycoproteins. Finally, upon maturation, the biofilm differentiates according to location within the biofilm community.

ANIM: Stop motion of bacteria landing and biofilm growth.

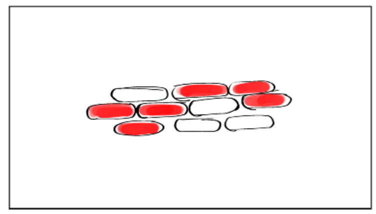
2-5



AUDIO: The most well-known form of biofilm communication is quorum sensing. This extracellular communication depends on the release of small chemical signals which coordinate bacterial behavior.

ANIM: Pan right and stop motion of quorum sensing.

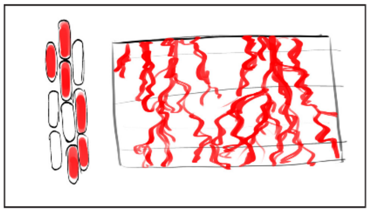
2-6



AUDIO: Scientific research cell to cell bacterial signaling dependent on potassium ions and their ion channels to communicate over long distances. Signals travel cell to cell, like oil through cracks in the earth, via percolation.

ANIM: Pan right and stop motion of bacterial electrochemical signaling.

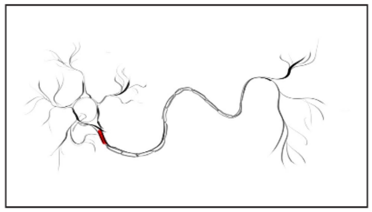
3-1



AUDIO: Scientific research cell to cell bacterial signaling dependent on potassium ions and their ion channels to communicate over long distances. Signals travel cell to cell, like oil through cracks in the earth, via percolation.

ANIM: Camera rotates 90 degrees and stop motion of percolation

3-2



AUDIO: This process is very similar to the signals which propagate along neurons, yet more slowly. Neuron signals travel along the length of the cell's axon.

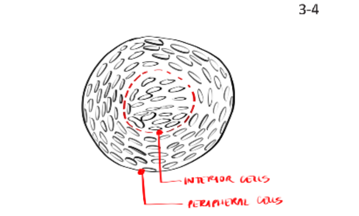
ANIM: Pan right and neuron signaling stop motion plays. Rotate 90 degrees back.

3-3

AUDIO: This new form of bacterial communication travels from cell to cell over a long distance. But why does this happen?

ANIM: Globe drawn to represent long distance.

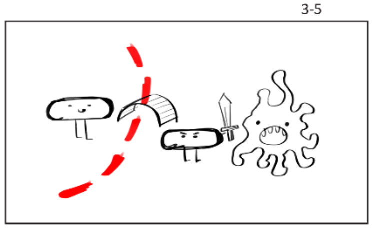
3-4



AUDIO: Biofilms have two distinct groups of cells: peripheral cells and interior cells. Biofilms have two distinct groups of cells: peripheral cells and interior cells.

ANIM: Biofilm showing interior and peripheral cells is drawn.

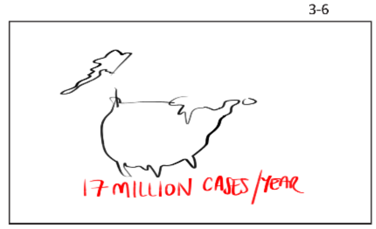
3-5



AUDIO: Peripheral cells have more access to nutrients and serve to protect the biofilm from external attack. Interior cells have limited access to nutrients but can survive and maintain biofilm growth if the peripheral cells are destroyed. Also, cellular location within the biofilm creates gradients of oxygen and nutrients, establishing a need for metabolic coordination and necessitating more sophisticated methods of communication.

ANIM: Scene depicting close-up peripheral and interior cells is drawn.

3-6

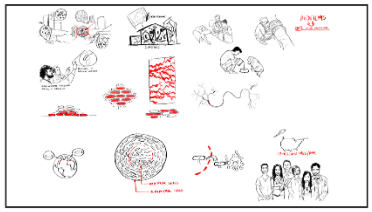


AUDIO: Over 17 million cases of biofilm associated infections occur every year in the US, resulting in over 500,000 deaths and an annual cost of $94 billion.
ANIM: Map and text drawn.

4-1

AUDIO: Further understanding of bacterial communication may hold the keys to both preventing and fighting these complex infections.
ANIM:Scene of diverse health professionals drawn.

4-2



AUDIO: ...
ANIM: Zoom out so that entire board is visible.

APPENDIX P

Exemption Granted

January 22, 2020

Sarah McGuinness
Biomedical and Health Information Sciences

RE: **Protocol # 2020-0081**
"Comparing Whiteboard and 3D Animation in Visualization of "Neuron-like" Bacterial Communication in Biofilms"

This research is approved for th enrollment of adult (18+ years) subjects only.

Dear Sarah McGuinness:

Your Claim of Exemption application was reviewed on **January 22, 2020** and it was determined that your research meets the criteria for exemption as defined in the U.S. Department of Health and Human Services Regulations for the Protection of Human Subjects [45 CFR 46.104(d)]. You may now begin your research.

Exemption Granted Date: January 22, 2020
Sponsor: None

The specific exemption categories under 45 CFR 46.104(d) are: 1, 2 and 3

You are reminded that investigators whose research involving human subjects is determined to be exempt from the federal regulations for the protection of human subjects still have responsibilities for the ethical conduct of the research under state law and UIC policy.

Please remember to:

- → Use your research protocol number (2020-0081) on any documents or correspondence with the IRB concerning your research protocol.
- → Review and comply with the policies of the UIC Human Subjects Protection Program (HSPP) and the guidance ***Investigator Responsibilities***.

We wish you the best as you conduct your research. If you have any questions or need further help, please contact me at (312) 355-2908 or the OPRS office at (312) 996-1711. Please send any correspondence about this protocol to OPRS via OPRS Live.

Sincerely,
Charles W. Hoehne, B.S., C.I.P.
Assistant Director, IRB #7
Office for the Protection of Research Subjects

cc: Anthony Faiola
Christine D. Young

Page 1 of 1

UNIVERSITY OF ILLINOIS AT CHICAGO
Office for the Protection of Research Subjects
201 AOB (MC 672)
1737 West Polk Street
Chicago, Illinois 60612
Phone (312) 996-1711

APPENDIX Q

Exemption Determination
Amendment to Research Protocol – Exempt Review
UIC Amendment #1

February 25, 2020

Sarah McGuinness
Biomedical and Health Information Sciences

RE: **Protocol # 2020-0081**
Comparing Whiteboard and 3D Animation in Visualization of "Neuron-like" Bacterial Communication in Biofilms

Please be reminded of the need to submit "marked" and "clean" copies of all revised documents with future amendments. Because this amendment did not include "marked" copies of the revised documents, the reviewers were required to conduct the review of this amendment as an initial Claim of Exemption application. This resulted in a delay of the review of the amendment and other investigators' submissions as well.

Dear Sarah McGuinness:

The amendment to your research has been reviewed. Your research continues to meet the criteria for exemption as defined in the U. S. Department of Health and Human Services Regulations for the Protection of Human Subjects [(45 CFR 46.104(d)].

The specific exemption categories under 45 CFR 46.104(d) are: 1, 2 and 3

You may now implement the amendment in your research.

Please note the following information about your approved amendment:

Amendment Approval Date: February 25, 2020
Amendment:
Summary: UIC Amendment #1: Involves changes to the recruitment materials, specifically:
1. Editorial revisions of the announcement made to classes;
2. The recruitment informational card has been removed from the recruitment materials; and
3. The recruitment method (how, where, when, and by whom) has been altered to accommodate recruitment of graduate students from UIC's College of Medicine and College of Dentistry.

You are reminded that investigators whose research involving human subjects is determined to be exempt from the federal regulations for the protection of human subjects still have responsibilities

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201 AOB (MC 672)
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Please remember to:

→ Use your research protocol number (2020-0081) on any documents or correspondence with the IRB concerning your research protocol.
→ Review and comply with the policies of the UIC Human Subjects Protection Program (HSPP) and the guidance ***Investigator Responsibilities***.

We wish you the best as you conduct your research. If you have any questions or need further help, please contact me at (312) 355-2908 or the OPRS office at (312) 996-1711. Please send any correspondence about this protocol to OPRS via OPRS Live.

Sincerely,
Charles W. Hoehne, B.S., C.I.P.
Assistant Director, IRB #7
Office for the Protection of Research Subjects

cc: Anthony Faiola
Christine D. Young

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1737 West Polk Street
Chicago, Illinois 60612
Phone (312) 996-1711

APPENDIX R

Exemption Determination
Amendment to Research Protocol – Exempt Review
UIC Amendment #2

February 25, 2020

Sarah McGuinness
Biomedical and Health Information Sciences

RE: **Protocol # 2020-0081**
Comparing Whiteboard and 3D Animation in Visualization of Neuron-like Bacterial Communication in Biofilms

Please refrain from submitting multiple amendments to the same research protocol at the same time. UIC Amendment #1 and UIC Amendment #2 could have easily been combined into a single amendment, which would have resulted in a faster and more efficient review.

Dear Sarah McGuinness:

The amendment to your research has been reviewed. Your research continues to meet the criteria for exemption as defined in the U. S. Department of Health and Human Services Regulations for the Protection of Human Subjects [(45 CFR 46.104(d)].

The specific exemption categories under 45 CFR 46.104(d) are: 1, 2 and 3

You may now implement the amendment in your research.

Please note the following information about your approved amendment:

Amendment Approval Date: February 25, 2020
Amendment:
Summary: UIC Amendment #2 involves the following:
1. The research title has been modified to remove the quotations from "Neuron-like" in order to fulfill the character limit for UIC thesis title requirements; and
2. Three questions (Survey Comparing Both Stimuli) have been modified to remove bias from the questions.

You are reminded that investigators whose research involving human subjects is determined to be exempt from the federal regulations for the protection of human subjects still have responsibilities for the ethical conduct of the research under state law and UIC policy.

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1737 West Polk Street
Chicago, Illinois 60612
Phone (312) 996-1711

for the ethical conduct of the research under state law and UIC policy.

Please remember to:

→ Use your research protocol number (2020-0081) on any documents or correspondence with the IRB concerning your research protocol.

→ Review and comply with the policies of the UIC Human Subjects Protection Program (HSPP) and the guidance ***Investigator Responsibilities***.

We wish you the best as you conduct your research. If you have any questions or need further help, please contact me at (312) 355-2908 or the OPRS office at (312) 996-1711. Please send any correspondence about this protocol to OPRS via OPRS Live.

Sincerely,
Charles W. Hoehne, B.S., C.I.P.
Assistant Director, IRB #7
Office for the Protection of Research Subjects

cc: Anthony Faiola
Christine D. Young

UNIVERSITY OF ILLINOIS AT CHICAGO
Office for the Protection of Research Subjects
201 AOB (MC 672)
1737 West Polk Street
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Phone (312) 996-1711

APPENDIX Q

See Supplementary Files for data sets.

VITA

NAME:	Sarah Fiona McGuinness
EDUCATION:	PhD Candidate in Bioengineering University of Illinois in Chicago, 2020 Computational Modeling of Proteins and Materials
	Master of Science in Biomedical Visualization University of Illinois in Chicago, 2018 to 2020 Thesis: Comparing Whiteboard and 3D Animation in Visualization of "Neuron-like" Bacterial Communication in Biofilms
	Bachelor of Science in Biochemistry and Genetics, Minor in Art Texas A&M University, 2013 to 2017 Thesis: In Situ Hybridization of Spleen Sections of Young Adult Female Rats Observed For Presence of miRNA-363-3p
PROFESSIONAL:	Teaching Assistant, College of Applied Health Sciences, AHS 375 Ethics Law in Health August 2019 – May 2020
	Graphic Designer, Department of Disability and Human Development, August – December 2019
	Environmental Monitoring Technician I, FUJIFILM Diosynth Biotechnologies, January 2018 – July 2018
	Student Researcher, Texas A&M Department of Soil and Crop Sciences PI: Dr. George Hodnett, Senior Research Associate, May - December 2017
	Texas A&M Health Science Center, Department of Neuroscience and Experimental Therapeutics PI: Dr. Amutha Selvamani, Associate Research Scientist, January 2016 - May 2017
	Texas A&M Center for Phage Technology PI: Dr. Jason Gill, January - May 2015
PROFESSIONAL MEMBERSHIP:	AMI (Association of Medical Illustrators) Student Member, 2019 – 2020
	SAMA (Student Association of Medical Artists) Executive Position: Social Media Chair, 2019 - 2020 General Member, 2018 - 2020

Texas A&M Town Hall
Executive Officer of Advertising, Marketing, and Promotion, 2014 - 2016
General Member, 2014 – 2016

EXHIBITS: SAMA Show 2020: Unique Perspectives: Art and Science in Medicine
International Museum of Surgical Science, January 2019

SAMA Show 2019: Capturing the Beauty Within
International Museum of Surgical Science, February 2019

PUBLICATIONS: The Northwestern Public Health Review Cover, Winter 2019 Issue
"The Immunological Principles of Allergies"