Mixed Methods Evaluation of Immuno-Defender's Player Experience and Learning Outcomes

by

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This study evaluated the prototype game *Immuno-Defender*, a 3D action game that teaches players about the human immune system. The pilot study assessed the feasibility of combining multiple qualitative and quantitative methods to evaluate a learning game. Six participants took part in a remote playtesting session with the investigator, which was book-ended with a pre and post test to assess knowledge acquisition and a self-assessment of player experience using a shortened version of the Player Experience Inventory (PXI). Participants then completed a delayed posttest to assess knowledge retention. Qualitative analysis included observation, retrospective, interview, and a player performance assessment. Results showed that participants increased their knowledge assessment scores ($t = 5.59$, $df = 5$, p-value of 0.002) and the four that completed the delayed posttest retained their knowledge ($t = 5.64$, $df = 3$, p-value of 0.011). Qualitative findings revealed the strengths and weaknesses of the game's current design and provided insights for the next iteration of development. Visit ajmrkva.com/immunodefender to learn more about the project.

Keywords: mixed methods, transformative games, serious games, educational games, videogames, digital games, games for health, playtesting, remote playtesting, player experience, games user research, immunology, immune system.

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Preface

I dedicate this to Nora, the righteous love of my life.

In loving memory of Gail Bridges.

Acknowledgements

None of this would have been possible without the generous support of so many people. Let me be crystal clear. I would have quit pursuing this Dragon several times if it were not for several key individuals that gifted me the courage to continue despite the overwhelming and discouraging obstacles in my path.

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I owe it to Dr. Grieve who first taught me game design and convinced me that making games was a dark art worth pursuing. My journey at this school started with us meeting at a Starbucks, and since then Dr. Grieve demonstrated the application of game design in teaching as well as opening questions like a series of doors; leading me deeper down the rabbit-hole.

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I owe it to Dr. Brusilovsky who lent a critical eye and constructive feedback in teaching me human-centered computing.

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Finally, I want to thank you, dear reader. If you are reading this thesis, then all the work to create it was worth it. Let me know what you think. I'm on the Internet.

1.0 Introduction

How humans survive diseases is both complex and fascinating. The human body has interconnected systems of natural defenses augmented with medical treatments including vaccines. Viruses and bacteria themselves have their own counter offenses and ways of overcoming our defenses. This ongoing microscopic battle impacts everyone, and yet many of us do not fully grasp what is happening including college students pursuing careers in pharmacy and biology.

Transformational games are designed with the intent to take such individuals and transform their knowledge and behavior beyond the game world in such a way that persists long after their experience playing the game has ended [15]. Serious games are a kind of Transformational game that utilizes game principles for learning, skill acquisition, and training purposes [34]. They have been extensively studied and recognized for their effectiveness ([16], [51]), and have gained popularity as a prominent tool in professional health education [12]. Among the many applications, serious games centered on the complex domain of the immune system have been shown to educate players of all ages while delivering an engaging experience [58].

While it's possible to design and develop a serious game, it is a difficult undertaking that requires creativity, iterative experimentation, research, time, and money. Games have been built over years by teams of domain experts, designers, and developers. Generally, these games begin as a prototype that undergoes a series of playtesting and evaluation studies to determine if they are reaching their purpose as serious games or need drastic rethinking. Limited production resources and the need to socially distance because of pandemics like COVID-19 continue the need to evaluate games remotely. Remote playtesting is also favorable for playtesters that want to be comfortable playing at home on their own devices, online multiplayer games, and for playtesters that have varying schedules and cannot participate during normal business hours.

In this thesis, I present a mixed methods research study to evaluate an early prototype of *Immuno-Defender*, a game I'm designing and developing under the guidance of my team

of mentors and experts (Figure 1). The games' primary goal is to be an immersive, fun, and engaging experience, while having the consequence of also teaching the player about the immune system by putting the immune system functions in their control. I decided to pursue this particular game to both explore the fascinating world of the human body's defense system and to educate not only students but also the general public about something as vital as the immune system. Furthermore, the ongoing distrust of vaccines and widespread misinformation about COVID-19 has motivated me to focus on games for public health, and aim for an audience of curious and passionate learners [63].

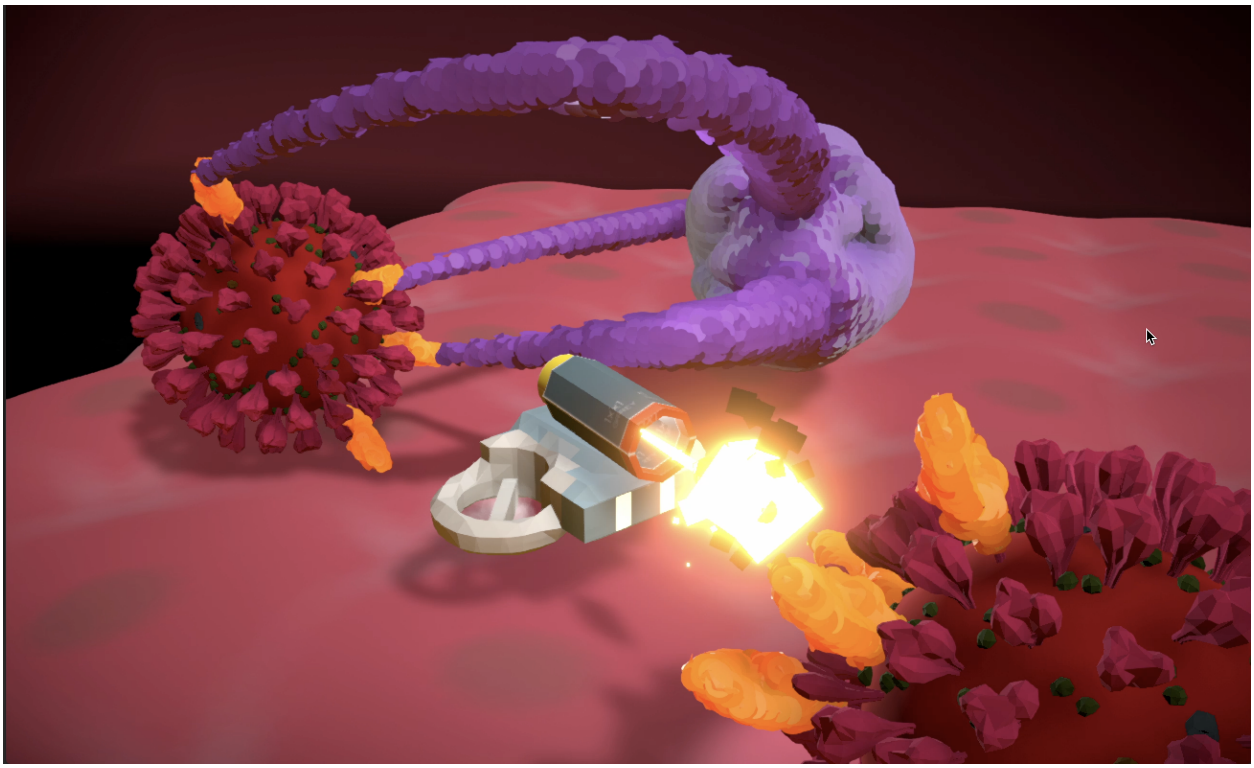


Figure 1: Defend against invading pathogens the immune system way.

I evaluated the prototype's player experience and knowledge acquisition potential by putting it in the hands of its target audience in a mixed methods pilot study with six participants. The project pilot-tested a methodology that combines remote playtesting, retrospectives, interviewing, player performance grading, and questionnaires (a recruitment

survey, pretests and posttests for knowledge acquisition, a delayed post test for knowledge retention, and a questionnaire for self-assessing player experience). The games industry has adopted remote playtesting and game design researchers have begun to test this approach that is flexible and economical for both researchers and participants [4]. The results of my study will contribute to game design research, serious game research, the use of games in immunology education, and help researchers interested in conducting small-scale remote studies of serious game prototypes.

2.0 Background

2.1 Immunology Curriculum

My goal was to create a game that could provide a gratifying experience to a novice learner in immunology and keep them engaged through increasingly complex gameplay and levels that dive deeper into the depths of immunology. Such a game would have potential to complement the immunology curriculum at the university level, particularly for students who benefit from active or game-based learning resources. Immunology is a difficult subject to learn given it covers complex systems overlaid with dense jargon and delivered in lectures that students "often report boredom and loss of interest after approximately 15 min" [33].

Dr. Kerry Empey is an Associate Professor in the Department of Pharmacy and Therapeutics and Center for Clinical Pharmaceutical Sciences. She described the pain points of teaching immunology to college students, which covers a complex system of interconnected parts performing multiple functions at various time points. Dr. Empey expressed that a game could facilitate learning immunology by visualizing the system and having the player perform many of the functions.

In addition to lectures, current pharmacy students learn from their assigned readings including the fifth edition of The Immune System [47]. They also have access to videos from the publisher (Norton) or assigned by the instructors covering the lessons. While the book and supplemental readings are informative and essential to learning, their format is restricted to text, tables, and images. The videos provide more visualization but are not interactive. All these resources were used in the design of the game *Immune-Defender*, which sets out to supplement these resources with something entertaining and interactive.

2.2 Leveling Up Education

Games excel at providing dynamic and immediate feedback to learners, adjusting difficulty to skill level, having "infinite patience," and most of all, making failure fun and rewarding [34]. This last trait is missing most in the classroom where failure penalizes students with lower grades and the curriculum moves forward instead of progressing when the student is ready. Games are another form of learning where failure is part of the design, encourages resiliency, and players stay engaged to overcome their own limits [28].

The immersive and engrossing nature of games also excel at teaching procedural knowledge as well as improving knowledge retention [56]. According to a meta-analysis of 79 empirical studies, "post-instruction learning achievement was weakly to moderately higher for declarative knowledge, knowledge retention, and procedural knowledge for students taught with serious games [versus instruction using more conventional methods]" [51]. Therefore, serious games hold the potential to teach the procedural knowledge of the immune system (not just what is in the system but how does the system work? How does a macrophage consume a pathogen and what is the complement system doing and when?). Games go beyond the barriers of margins, footnotes, and tables by visually simulating in motion and function an interconnected network of actors and elements.

Digital games have some advantages over analog games, particularly for evaluating and improving learning outcomes. Digital games can collect data on the player's actions and decisions and share that data with teachers, researchers, or use the data to dynamically respond with helpful tips or adjust the difficulty. In fact, a thoughtfully designed game can perform a stealth assessment of players' learning by having the assessments integrated so well into the game design that the player can solely focus on playing the game and be unencumbered from the interruptions (or anxiety) of surveys and questionnaires [55].

2.3 Why *Immuno-Defender*?

Given the importance of the immune system, its complexity, and the ability for serious games to embrace complexity, there have been a number of immune system games and some of them have been part of studies. The main focus of this work has been to evaluate the knowledge acquisition value of immune system games. Steinman, et al. at the University of Pittsburgh found their trading card game to be effective at teaching or for practice depending on the age of the participant [58]. *Cells of War* was a digital card game that was evaluated in a pilot study to gather feedback for further development [36]. Cheng et al. compared the effects of their 2D game, *Humunology*, to web-based curriculum to find that students that played the game "outperformed those who learned by using web-based content..." [11]. Stegman found that students that played her game, *Immune Attack*, outperformed students that played a control game and were more confident in understanding advanced textbook visuals [57].

While there is a growing corpus of games designed to educate players about the human immune system designed to educate players, there are gaps in what is available and only a drop in the ocean of what is possible. Most of the games discovered during the lit search were not available to play. In fact, my literature search found an alarming amount of serious games in published studies that were simply not available to purchase or play including *Steinman & Blastos*' card game, *Cells of War*, and *Humunology*. If players cannot access games that once filled a serious game niche, then there is an opportunity for players to enjoy and learn from a new game.

I found no studies that explored why so many serious games seem to be limited in distribution or become vaporware, but I personally value games that are visually rich, immersive, challenging, and invoke feelings like excitement and curiosity. That is why I paid close attention to the only serious games about the immune system that came close to my gaming preferences and were still playable: *Immune Attack* and *ImmuneQuest* ([49], [57]). While *Immune Attack* is a fully formed 3D game where players learn about the immune system by exploring it in a nanobot (in an almost identical description to one of my early concepts), the game in its current form feels outdated and is not very fun to play in my testing using

the Spring of 2008 build.

ImmuneQuest is visually more up-to-date and provided an excellent exchange of information bites to player actions throughout gameplay, but while it executes well on the real-time strategy (RTS) genre, it became either too slow or not challenging enough to sustain my interest or invoke feelings during my tests. There are still more games to explore in the immune system domain, but from my research I have identified the niche I am designing to fulfill and now have a prototype to test it.

3.0 Game Design and Development

The game design and development process may look different from one indie developer to another or one game studio to another, but it generally follows the standard software development cycle. The first stage is conceptualization where the game concept is chosen. This is followed by the creation of a prototype (paper prototypes are best to begin with but in my case I have a digital prototype). Next, playtesting the prototype takes place. Evaluating the results of playtesting is then followed by a return to conceptualization and the next round of design and prototyping. Figure 2 is a simple reproduction of this process' diagram as found in the book Game, Design, and Play: A Detailed Approach to Iterative Game Design [41].

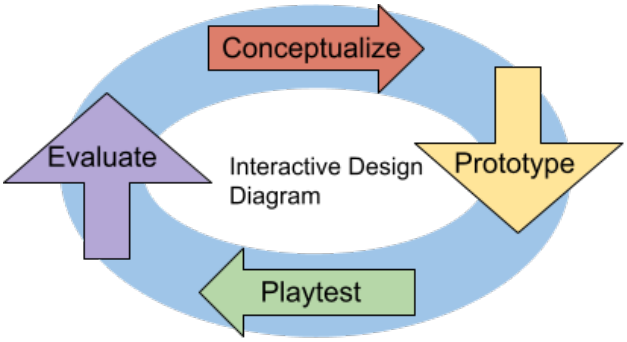


Figure 2: Iterative Game Design Process

3.1 From Concept to Playable Prototype

Many hours of learning and experience led to creating the current prototype of *Immuno-Defender*. As a part-time student working full-time at the University of Pittsburgh (first as a Data Manager and now as a Data Analyst), I completed a series of classes on game design and development at the University of Pittsburgh. I also completed multiple independent studies with my mentors Dr. Babichenko, Dr. Grieve, and Dr. Patel that explored game

design and development while also participating in Game Jams that furthered my experience with ideation and prototyping. One of those projects was an educational tower defense game about antibiotics where the player set up defense towers that represented different antibiotics to combat a infection from different pathogens (Figure 3)[2]. During 2021, I began to pivot towards a game about the immune system that I would design, develop, and evaluate.

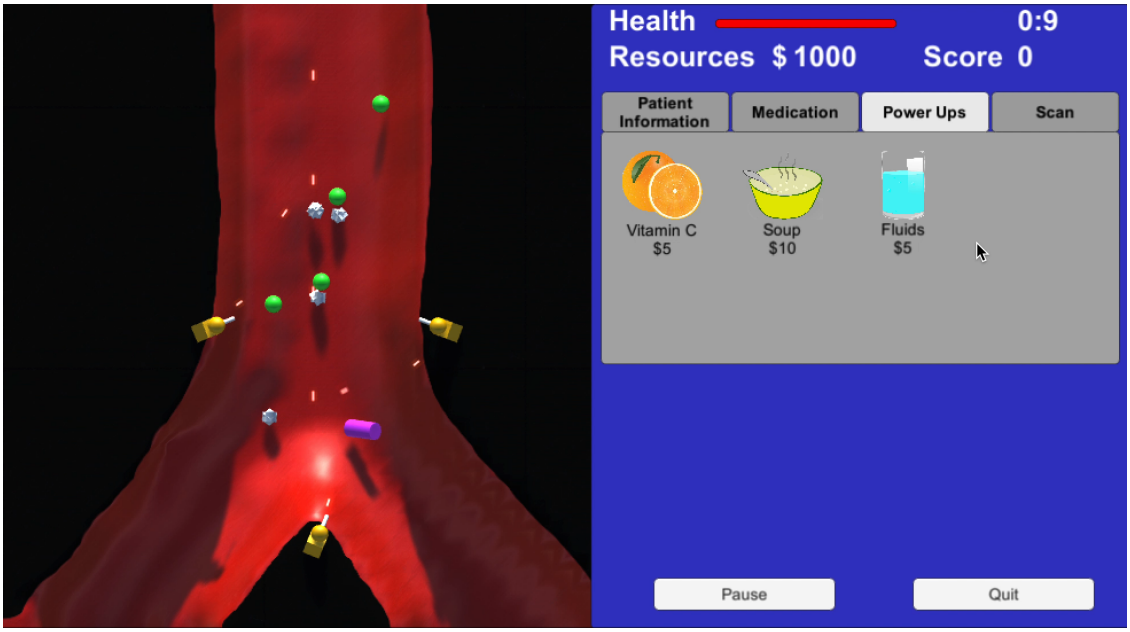
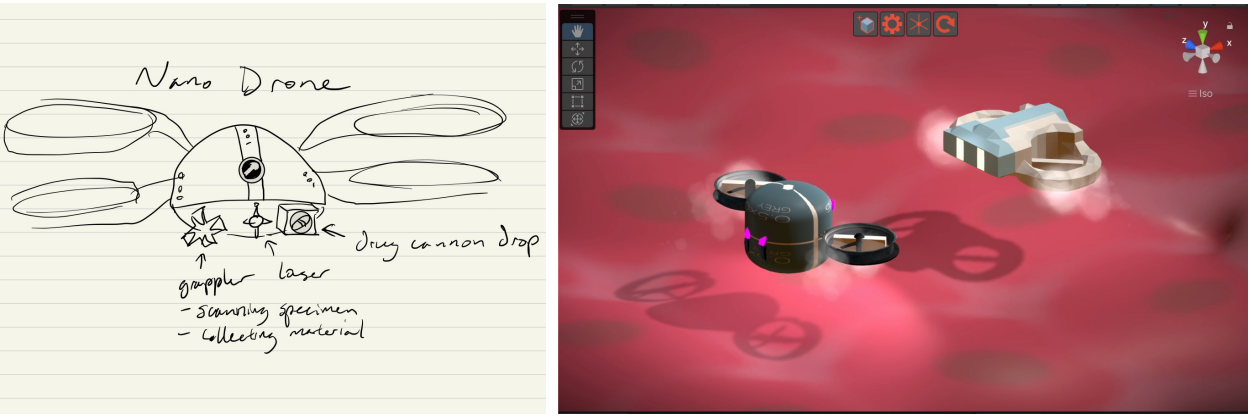


Figure 3: Body Defense Prototype

Early in 2021, I began exploring game concepts that would put the player in the middle of immune system functions. I began learning more about the innate and adaptive immune system and found potential mechanics, visuals, objectives, and conflicts to explore in a game. My research of related games (and playing the ones that are still accessible) gave me an idea of what has been achieved and what was missing. After several conceptual iterations, I settled on a design for a 3D third-person game where the player controlled a NanoDrone (tiny remote controlled robot like in Figure 4) that could explore inside the human body and interact with cells and pathogens.



(a) NanoDrone Concept (b) Early and Late NanoDrone Models

Figure 4: NanoDrone Design Iterations

I designed *Immuno-Defender* to put the player at the center of the immune system in a 3D fast paced immersive experience. Immersion in this case refers to the player experiencing a state of flow where the player is intensely focused on the task to the point of losing the sense of time and surroundings [14]. Balancing accessibility for a wider audience, immersion, and the need for minimal art assets to reduce production costs also led to switching the games' perspective from a 3rd-person player controlled camera to a top-down camera that follows the player. Figure 5 demonstrates the top-down perspective where the camera follows the player's character in the 3D environment.

Long term goals guided the design and development of the prototype. The game's architecture was designed to expand with new levels, additional layers of the immune system, and cooperative multiplayer if playtesting proves the game is viable. Expanding the game into a multiplayer experience would be highly useful in a classroom setting and a feature that is not seen in many learning games (especially games about the immune system).

With these core design decisions made, iterative design and prototyping led to developing a series of core game mechanics for the playable prototype. Between reading The Immune System book [47] and discussing ideas with Dr. Empey, Dr. Grieve, and Dr. Patel, I developed a core gameplay loop focused on the innate immune system responding to and defeating an influenza viral infection. In the game, the player first uses the complement

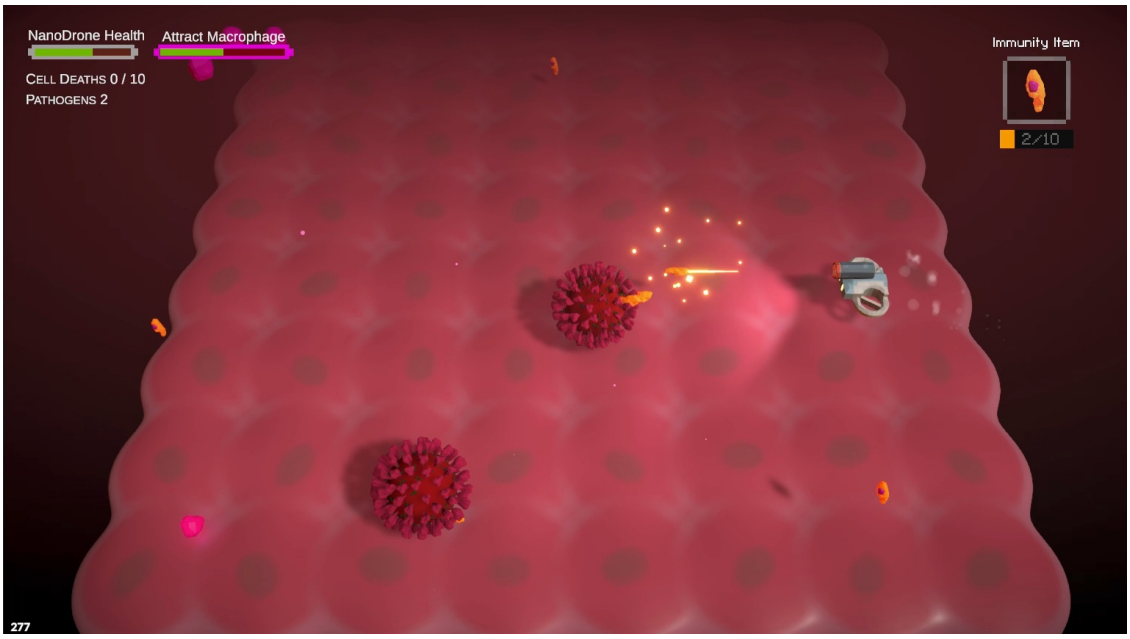


Figure 5: Top-down perspective of complement combat system.

system to mark the pathogen as a threat, then attracts a macrophage, then controls the macrophage to eat the pathogen, and repeat until they are successful. I then redeveloped the inflammation feature after discussing its role with Dr. Grieve, and added a new immune cell–the M2 Macrophage–after learning about it from Dr. Empey. In the discussion chapter I will elaborate on these features and more having analyzed their impact on the player experience and learning objectives.

Immuno-Defender started with a concept about an immersive and fun game about the immune system. Conceptualization included research, discussions with domain experts, designs, redesigns, and eventually led to a series of rough prototypes to experiment with different concepts before arriving at a prototype worth playtesting with people outside myself and my network. Playtesting is the essential next step in the development of *Immuno-Defender*. "The playtesting moment in the iterative cycle is where you find out the truth about your game's design" ([41], p. 201).

4.0 Research Design and Methodology

The act of playing a game is a dynamic experiential event where a player must learn the rules, procedures, and objective of the game through systems of audiovisual communication, all the while being cognitively and emotionally connected to the game. Playtesting puts the experience under a microscope in order to understand what the player is thinking, feeling, and what needs to change in the design for this experience to be as intended. Playtesting a learning game adds the need to understand what the player is learning and what the game is failing to teach them. For my study, I have come up with five research questions to answer by analyzing playtests of *Immuno-Defender*. I also have seven specific aims that defined the design of my methodology and analysis.

Research Questions

RQ1. What is the player experience of *Immuno-Defender*, as determined by analyzing remote playtesting sessions and questionnaires?

RQ2. To what degree does playing *Immuno-Defender* impact knowledge acquisition regarding actors and their functions of the innate immune system?

RQ3. To what degree does the participant retain their knowledge after three weeks following their first post-test?

RQ4. To what degree does a participant's player experience, as captured quantitatively by their Player Experience Inventory answers, correlate to their score on a test that assesses their knowledge of immune system functions.

RQ5. To what degree does a participant's in-game performance of *Immuno-Defender* correlate to their score on a test that assesses their knowledge of immune system functions?

Specific Aims

Aim1. Describe the target audiences' gaming habits, preferences, and interest in games (or more specifically videogames about immunology) based on the participant survey results.

Aim2. Evaluate participants' acceptance, engagement, and level of immersion of *Immuno-Defender* by combining the qualitative analysis of remote playtest sessions and quantitative analysis of their Player Experience Inventory results.

Aim3. Analyze data to identify strengths, weaknesses, new ideas, and ways to improve the game.

Aim4. Assess viability of the game's design and learning objectives for deciding to continue development or change direction.

Aim5. Assess the feasibility of combining remote playtesting, game immersion assessment, and knowledge acquisition assessment in the same study.

Aim6. Assess the effect of *Immuno-Defender* on participants' knowledge and knowledge retention of immune system functions in the context of a pilot study.

Aim7. Assess the relationship between player experience as measured by the Player Experience Inventory and learning outcomes.

4.1 Target Audience Research & Playertester Profiles

The first phase of the study began with a recruitment survey (Appendix A.2). To enroll in the study, the participants first completed a Qualtrics Survey[1] that included screening questions, questions about gaming preferences, questions to measure their interest in learning and playing games about the immune system, and basic demographics. This data provided context to the player behavior and feedback collected in the playtests.

Each participant that completed the study was given a unique ID to ensure confidentiality (e.g. P1 being the first participant). I will be referring to this unique ID throughout the thesis to tie together the different results in my analysis. Certain results will be relevant to a specific findings (such as an Expert gamer expecting certain keyboard controls the game doesn't support, or a Casual gamer not recognizing the health bar in the top-left corner

[1]Qualtrics is an online survey service that University of Pittsburgh staff, faculty, and students have access to utilize in research endeavors [48]

of the screen), which is why the recruitment survey included a breadth of questions about gaming experiences and preferences in order to create playtester profiles.

For the player to experience the game as intended, they need to learn the game's rules and procedures. Over the years, designers and players have constructed a lexicon of design standards and expectations, with ever more granular expectations within specific genres. From keeping points in a game of Rummy to having a Health Bar in an action game, players learn these features early on and recognize them–if not expect them outright. Players that have experience playing games and specific genres have built up a mental model of what to expect and what they prefer. Game designers can leverage these affordances and preferences to create more complex and daring games that would not be approachable to new members of the gaming community.

When it comes to playtesting, a player with extensive experience playing games will have that experience to draw on when playing the prototype and provide feedback. This can also mean that they have certain expectations of how to play a game based on what they have learned in the past, or if they are new to games they will not be familiar with the affordances and possibly struggle. Having some idea of the player's experience level and preferences helps the investigator to interpret their behavior and feedback in the playtest. The recruitment survey included a section to ask respondents a series of questions about their gaming experience and preferences. For example, this particular prototype is a 3D single-player action game that requires the player to use a keyboard and mouse, and thus includes questions about experience and preference for perspectives (2D, 3D, etc.), single or multiplayer games, genres, and input devices.

4.2 Qualitative Evaluation of Player Experience

The core task of the evaluation is to playtest the prototype for crucial feedback. Sample size for playtesting can vary based on the stage of development, scope of the game, and budget. Because this study playtests a prototype, usability testing, where five participants can find 85% of the usability problems in a prototype [44]. Winn and Heeter recruited six

playtesters for their target audience whey they playtested their game *Life Preservers* [65]. I recruited six participants in order to evaluate the prototype without wasting valuable time and resources by sampling more than is needed for an early prototype.

Each participant received instructions and completed a pretest before attending their playtest session (Appendix A.3). I scheduled and recorded playtesting sessions using Zoom. During the session, I guided the participant to download and play the game. During the start of the session, I observed the participant play as they thought aloud describing their actions. I also used a semi-structured interview guide to further probe for feedback and follow up on the participant's interpretations and actions in the game (Appendix A.4). During the wrap-up of the playtest, participants were reminded to complete their posttest, which included questions from the Player Experience Inventory as well as human immune system questions (Appendix 7). The screen and audio recording supplemented notetaking for qualitative analysis.

4.3 Declarative & Procedural Knowledge Acquisition & Retention

While the prototype was not developed enough to warrant a full study of knowledge acquisition (too much of the game can change between this iteration and the next and significant design or technical issues can interfere with learning outcomes), this pilot study included several methods for evaluating how the prototype potentially impacts the learning outcomes it is intended to influence. In order to measure the game's effect on a participant's declarative knowledge of the immune system response as well as their retention of the knowledge, participants completed a pretest before the playtest session and a posttest afterwards that included a series of questions about the immune system, as well as ask the participant's comfort level with their answers and to describe which questions they found most difficult (Appendix A.3, 7). Out of the eleven questions in the Qualtrics survey, seven were multiple choice questions I obtained from the W.W. Norton and Company Publishers Test Bank, which is available to instructors deploying their tools for creating curriculum alongside the book The Immune System [47]. The other four questions were open-ended questions to cover

features not captured in the Test Bank.

The same questions were used in all three tests and were randomly ordered for each participant at each time-point. I distributed the delayed posttest three weeks after the playtest to test their knowledge retention; a duration demonstrated in Delasobera's study [17] (Appendix A.6). The delayed posttest also asked if the player had played the game since the previous posttest, and to share their thoughts about the game if they had.

I was not able to calculate the Cronbach's alpha for the tests as they contained some missing data since participants were allowed to skip questions if they did not know the answer. When scoring the answers, participants received a half point if they chose a similar answer (C3A instead of C3B, epithelium instead of endothelium) or were partially correct in the open-ended questions. For analysis, I removed one question when I determined it was not actually part of the learning objectives.

To evaluate procedural knowledge, I drew on the work of Sitzmann who defined evaluating procedural outcomes in her 2011 article, "Procedural outcomes were defined as the ability to perform the skills taught in training. They were assessed by participating in an activity (e.g., simulation or role-play) or with a written test that required trainees to demonstrate memory of the steps required to complete the skills taught in training."[56] My approach included both recalling the steps of the innate immune system as well as a skills-based test of performing those steps.

To measure the game's effect on a participant's procedural knowledge of the immune system response, participants were graded on their recall of the immune system process as well as their performance in applying that knowledge to defeat the pathogens in the game (). The interview stage of the playtest session begins with a retrospective where I ask the player, "What happened in the game?" A retrospective is a useful method in games user research to unpack the player's comprehension of the experience [20]. My implementation is to both understand the player's interpretation of what happened in the game and to grade what immune system elements they include in their recall and whether they accurately describe their function in connection to the innate system procedure.

To evaluate how a participant applies their procedural knowledge in the game, I developed a rubric to grade each player's in-game performance at the last play-through (Appendix

A.7). My goal is have the player's performance depend on their procedural knowledge of the innate immune system. The original plan was to utilize a framework like Unity Analytics or Pat Healy's OpenGameAnalytics to capture in-game events, but I discovered through preliminary playtesting that the game was still too buggy for an automated performance system [61][31]. Instead, a qualitative and manual approach was needed to define not only how players perform but also what affects their performance.

4.4 Player Experience Self-Assessment

Having the researcher present during playtesting provided a rich volume of information from observing, listening, and discussing the participant's experience. It also carried the risk of various biases in the data collection. Observer bias can influence the researcher's notetaking and probes [38]. Another risk is the Hawthorne effect where the player will change their behavior because they are being observed and part of a study [23]. Additionally, some very key data points including assessing the participant's feelings and emotions during playtesting may be difficult in the remote setting.

To provide an alternative data point on player experience as a check against these risks, participants completed a simplified version of the Player Experience Inventory (PXI) alongside the posttest questions (Appendix 7) after the playtest session [3]. This validated questionnaire is designed to benchmark games based on the Mechanics-Dynamics-Aesthetics (MDA) framework, which formalizes measuring the player experience using a series of likert scale questions (Figure 6). The goal of the analysis is to supplement the qualitative findings as well as compare the results with the learning outcomes and player performance.

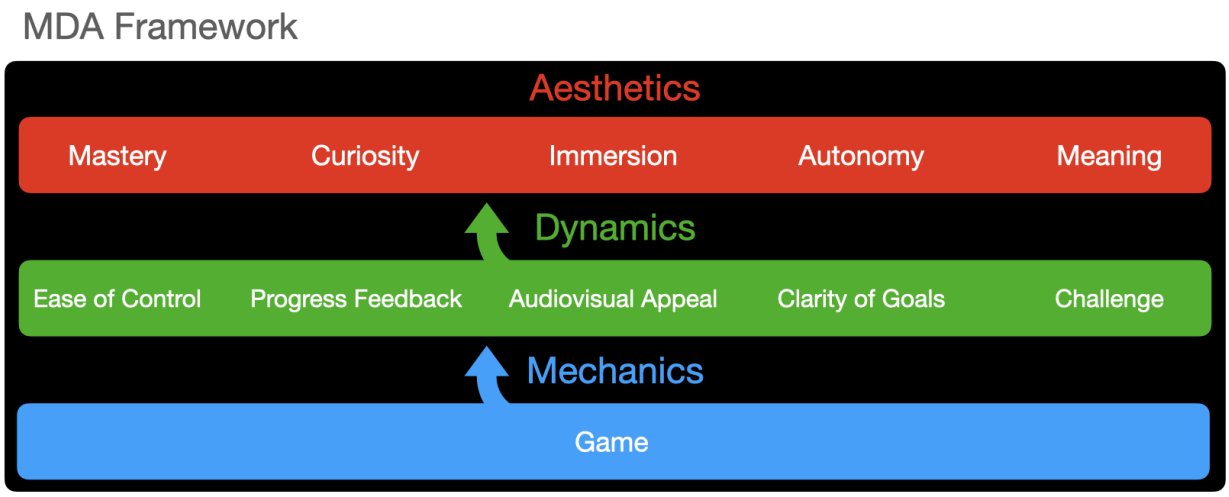


Figure 6: PXI construct based on MDA framework. [30]

4.5 Recruitment

Inclusion Criteria

- Participants must be 18 years of age or older.
- Participants must be undergraduate students who have not completed university level courses on immunology.
- Participants must have access and be able to use a computer, keyboard, and mouse.

Exclusion Criteria

- Participants with a history of migraines or epileptic seizures in response to visual or auditory stimuli will be excluded.
- Participants who have completed university-level immunology courses will be excluded.

The recruitment effort included requests through faculty, contacting presidents of student organizations, and posting fliers (Appendix A.1). This outreach directed students to an online survey where they decided if they wanted to participate and share their contact information. Effort was made to recruit a diverse demographic of the student cohort to match the demographic distribution of the school, but recruitment was also based on a

convenience sample that may not have produced a diverse outcome. There are no studies that indicate a difference in learning outcomes based on gender, sexual orientation, and race.

4.6 Data Analysis Methods

I explored the recruitment survey data to define the characteristics of respondents that completed the study and those that took the survey but did not complete the study. I described in the results any discerning differences between the handful that completed the study and the few that did not enroll. I then created a table to define each participant's gaming profile to contextualize their behavior and feedback in the playtest (Table 2).

For within subjects analysis of the pretest and posttest results, the Shapiro-Wilk test found normality between scores. I used the paired sample t-test to measure knowledge acquisition between the pretests and posttests within subjects. I interpreted the results of the analysis in the context of the small sample size for the purpose of assessing potential knowledge acquisition and retention. I also used the results of the quantitative analysis to supplement the qualitative findings.

I analyzed the PXI results to assess the strengths and weaknesses of the game's design in terms of the MDA framework. I further analyzed the results by comparing a composite of participant scores against the change in their pre-post test results to determine if there was a correlation in player experience and testing outcome using Pearson's correlation coefficient.

I gained an understanding of the player experience and uncovered patterns among the qualitative data using deductive and inductive thematic analysis [13]. This process considered all the playtesting notes, screen recordings, and distilled the key recommendations of how to improve the game, learning outcomes, the study design, and some other findings. Methods outlined in the literature on playtesting sessions guided playtesting procedure and analysis ([25]; [27]; [41]; [52]; [20]). These methods included not leading the player or giving them hints (instead, the investigator's role is to observe and take notes) and don't just watch what the player is doing but also try to read the player's emotions. I performed deductive

and inductive thematic analysis of the qualitative data using Descript[2] to transcribe the playtest sessions and Obsidian[3] to synthesize the coded excerpts into themes.

To evaluate both declarative and procedural knowledge together, I visually examined the retrospective scores for each player and learning objective against their pretest and posttest scores to find patterns between declarative knowledge scores and procedural knowledge scores. Patterns found in this visualization are mentioned throughout the discussion of learning outcomes.

For procedural knowledge and player performance, I first visualized the player performance as a heat map to explore what skills players excelled in or struggled in (except instead of cool and warm colors, I used bronze, silver, and gold). I also manually calculated the length of time each participant played the game by reviewing the screen recordings and displayed this data as a bar graph. I then used Tukey's rule to identify outliers in the change of test scores before reporting Pearson's correlation coefficient with and without outliers of the player performance scores and the change in the players' test scores [35].

[2]Descript is an audio and video editing software that enables the user to transcribe and edit text in sync with video files [19].

[3]Obsidian is a note-taking and knowledge-management software that enables the user to create an interconnected system of notes [39]

5.0 Results

5.1 Recruitment Results

I kicked off recruitment at the end of January 2023, when I sent my network of four faculty from the School of Pharmacy a recruitment email containing the survey link and they proceeded to share it with students. Having only one playtester after one month of recruiting, I modified the IRB and expanded inclusion criteria to all University of Pittsburgh students. I then emailed my advisor, Dr. Babichenko, the recruitment email and flyer who then distributed them to his students at the School of Computing and Information (Appendix A.1).

I continued recruiting until the end of April 2023, when I reached a total of six playtest sessions. By then eleven respondents filled out the survey including one non-student (whom was then ineligible as the study required student status). None of the respondents had played an immune system game before. Of the ten students, four were from the Pharmacy School and the other six were from the School of Computing and Information (SCI). Only one of the Pharmacy students enrolled and completed the study, the other three either enrolled but were lost to follow up or did not enroll. Five SCI students enrolled and completed most of the study (two did not complete the delayed posttest). The demographics of the six enrolled participants includes ages between 19-29 (median 22.5) and a mix of non-binary, female, and male students. Reviewing the results of the survey, we can find some differences between the students that enrolled and the students that did not.

Table 1: Enrollees and Non-Enrollees

				Human Immune System		
Status	Program	School	Play Games	Knowledge	Interested to Learn More About HIS	Interested to Play HIS Game
Enrolled	Undergraduate	SCI	Yes	Intermediate	Yes	Slightly
Enrolled	Undergraduate	SCI	Yes	Beginner	No	Moderately
Enrolled	Undergraduate	Pharmacy	Yes	Beginner	Yes	Extremely
Enrolled	Graduate	SCI	Yes	Advanced	Yes	Extremely
Enrolled	Graduate	SCI	Yes	Intermediate	Yes	Extremely
Enrolled	Graduate	SCI	Yes	Novice	Yes	Moderately
Not Enrolled	Graduate	Pharmacy	No	Novice	Yes	Very
Not Enrolled	Graduate	Pharmacy	No	Novice	Yes	Slightly
Lost to Follow Up	Other	Pharmacy	Yes	Novice	Yes	Very
Lost to Follow Up	Graduate	SCI	Yes	Advanced	Yes	Very

Some constants that are not included in Table 1: none of the students had taken classes on the subject of the immune system and none of the students have played games about the immune system. All the non-enrollees considered themselves to be "Novice: I know a little about the immune system" while the enrollees were more varied from Beginner to Advanced 1). All non-enrollees were interested to learn about the immune system, and all but 1 of the enrollees did as well. Interest in playing games about the immune system varied from slightly interested to extremely interested across groups. All students except for two play games, and those two did not enroll. There is a chance that the two non-gamers did not enroll because they do not play games.

One survey question asked about the learning style preferences of respondents based on the VARK model [37]. How the learning objectives were implemented in the game favor certain learning styles (for instance, how the game teaches phagocytosis is both visual and kinesthetic in that the player embodies the macrophage performing phagocytosis and watches the process unfold). Then there are learning objectives that are completely text-based and may favor the reading/writing learning style. While the game may favor solitary learners (since it is single-player), it does not favor social/interpersonal learners and auditory learners. Figure 7 visualizes each student's preference; showing that they have multiple preferences (including one enrollee that prefers all of them). Among the enrolled students, four students prefer both visual and kinesthetic learning styles while two did not, but did prefer learning through reading and writing. We can include this as a data point to compare to each

participant's learning preference and their learning outcomes.

Table 2 displays the relevant gaming attributes of the enrolled students. As you can see in the table, most participants were more experienced while two considered themselves to be casual gamers. Only one student answered that they had no experience with 3D games, but also listed two 3D games as their most played and favorite games. So I would reclassify them as very or extremely experienced with 3D games. All but one participant preferred the mouse and keyboard, and four out of six preferred the Action game genre. Two participants slightly preferred single-player games while the rest moderately or extremely preferred them.

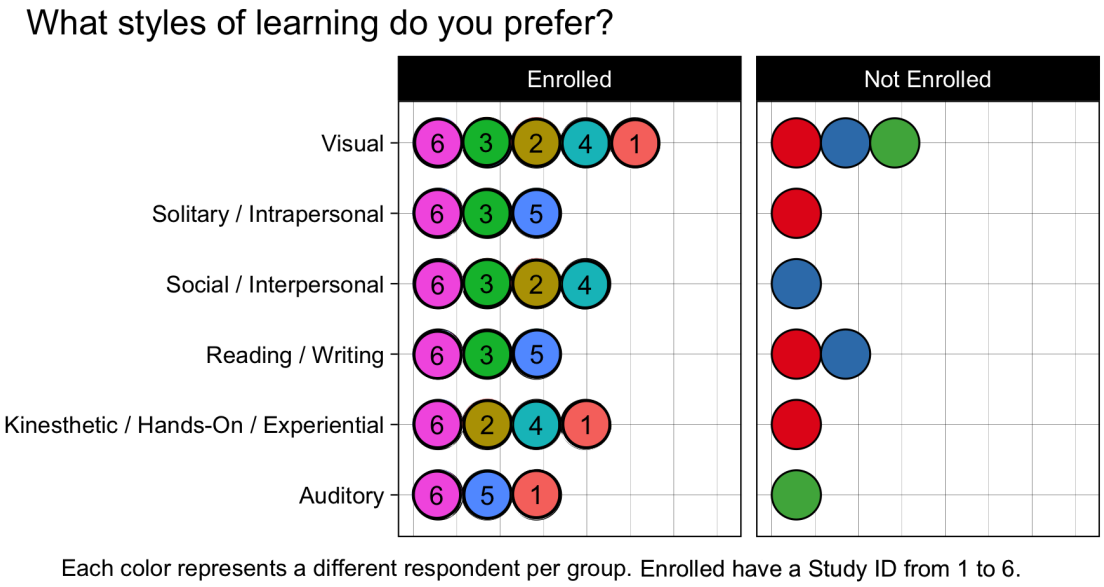


Figure 7: Learning Style Preference Per Respondent

Table 2: Gaming Profiles of Enrolled Students

					Player Preference		
PID	Gaming	Playtime	Frequency	3D Experience	Single-Player	Keyboard+Mouse	Action Genre
P1	Expert	1hr-<3hr	Weekly	Extremely	Extremely	✓	
P2	Expert	30m-<1hr	Daily	Very	Moderately	✓	✓
P3	Experienced	3hr+	Weekly	Very	Extremely	✓	✓
P4	Casual	<30m	Monthly	Very	Slightly		✓
P5	Casual	30m-<1hr	Weekly	Very	Extremely	✓	
P6	Experienced	30m-<1hr	Monthly	Not at all[a]	Slightly	✓	✓

[a]Participant listed 3D games as their favorite and most played games.

5.2 Knowledge Acquisition & Retention Results

RQ2: To what degree does playing Immuno-Defender impact knowledge acquisition regarding actors and their functions of the innate immune system?

5.2.1 Pretest, Posttest, Delayed Posttest

Using G*Power[1], for a one-tailed matched pairs t-test to compare the difference between two dependent means (pre and post, or pre and delayed, or post and delayed) using an effect size of 0.5 and a power of 0.8, I calculated needing 27 playtesters to achieve statistical power [8]. Even though 27 participants are required to achieve statistical power, the goal of this study is not to show that the learning outcomes will generalize to any immunology classroom, but rather to evaluate the current game's design in order to inform the next iteration of development and to pilot the study's methodology to inform the the design of a larger study needed for statistical power.

Game development is a long and expensive pursuit, which is why playtests are needed early on to test the game's design and implementation [18]. The same goes for testing the game in terms of learning objectives. The results of the tests cannot be generalized, but it gives the designer some evidence of whether the design has some impact. It is better than taking the gamble that the design will work down the road and finding out it isn't even close and that major costly redesigns are necessary. It also benefits the designer to implement and pilot the knowledge acquisition tests early in the process in order to fine-tune both how the tests are included in playtesting and to test the instruments themselves for accuracy. As we will see in the results below, when combining the test results with the qualitative data, the instrument does need further tuning for future iterations.

To answer RQ2, I start by defining my hypothesis:

H1: Playing *Immuno-Defender* increases the player's knowledge assessment score.

H0: Playing *Immuno-Defender* has no affect on the player's knowledge assessment score.

What statistical analysis I use to compare test results depends on whether the data is

[1]G*Power is a free software that enables researchers to compute a meriad of statistical power analyses and effect sizes.

normally distributed [53]. Calculating the Shapiro-Wilk normality test resulted in the p-values of the pre (W = 0.91, p-value = 0.46), post (W = 0.83, p-value = 0.13), and delayed post test (W = 0.91, p-value = 0.49) being all greater than 0.05, so we do not reject the null hypothesis and conclude that the data is normally distributed.

Calculating the paired sample t-test to measure knowledge acquisition between the pretests and posttests within subjects resulted in a p-value of 0.002. Meaning we can reject the null hypothesis and conclude that the change in score (t = 5.59, df = 5) was not random.
RQ3: To what degree does the participant retain their knowledge after three weeks following their first post-test?
H1: Changes in the player's knowledge assessment score after playing *Immuno-Defender* is retained after three weeks or more.
H0: Changes in the player's knowledge assessment score after playing *Immuno-Defender* is lost after three weeks or more.

Four out of the six respondents completed the delayed posttest. Calculating the paired sample t-test to measure knowledge acquisition between the pretests and delayed posttests within subjects resulted in a p-value of 0.011. Meaning we can reject the null hypothesis and conclude that the change in score was not random (t = 5.64, df = 3).

We conclude that the participants acquired some knowledge from the baseline pretest (mean 27%, sd 0.25) to the posttest after playing the game (mean 53%, sd 0.26), and retained most of that knowledge by the delayed posttest three or more weeks later (mean 71%, sd 0.23) (Figure 8). In fact, the delayed posttest score was higher but not significantly. P1 completed their delayed posttest ten weeks after the posttest and maintained their posttest score (75%) after all that time had past. P6 actually increased their delayed posttest score by 20% and stated that they did play the game between the posttest and the delayed posttest.

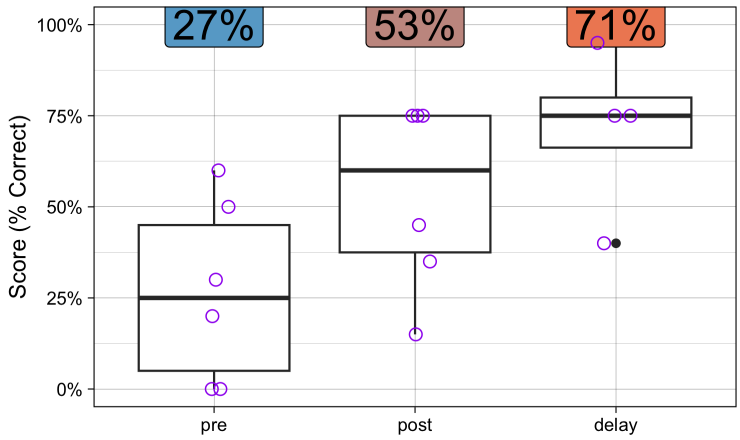


Figure 8: Overall Participant Test Scores

The pretests revealed that participants had a range of prior knowledge about the immune system. Figure 9 summarizes each participants score at each timepoint, and includes the percent of change between each point. Two participants scored between 50-60% on the pretest, whom I would identify as immunology hobbyists as both explained in the interview that they were interested in microbiology including the immune system and had absorbed content including videos on the topic. The third participant may not be a hobbyist, but was able to recall watching an episode of the Magic School Bus that explored the immune system and scored 30% on the pretest.

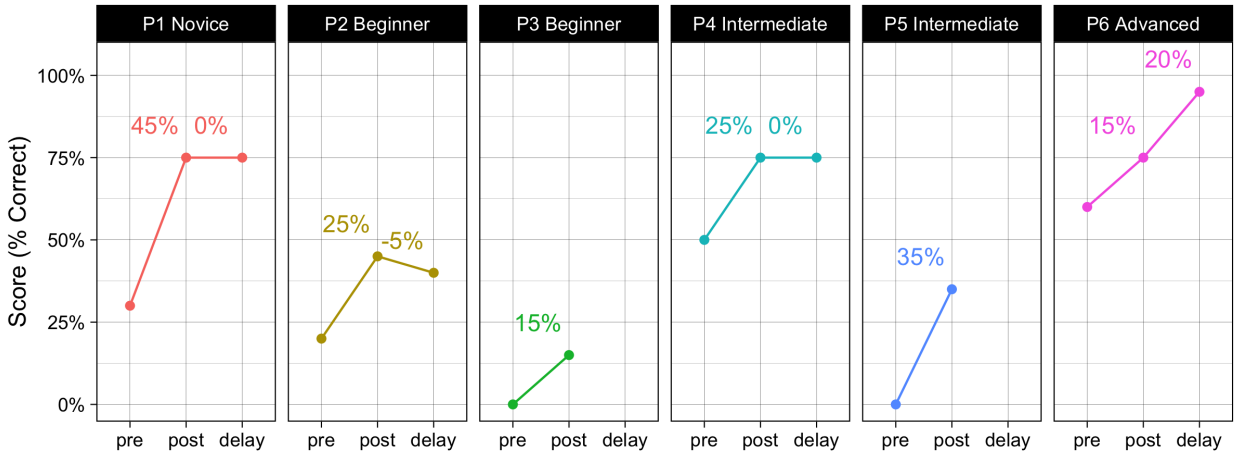


Figure 9: Participant score changes and self-determined immune system knowledge.

The other participants had lower pretest scores. One participant fell in the middle with a pretest score of 20% and was the sole pharmacy student that completed the study. The last two participants scored 0% on the pretest, with one answering "No" to whether they were interested in learning more about the immune system (and was the only participant not interested).

In the delayed posttest questionnaire, participants were asked if they had played the game since the previous posttest. P4 and P6 (the immune system hobbyists) said they had played the game after the posttest, and notably P6 increased their score to almost get a perfect delayed posttest score. It has the appearance that P6 could have played more to score higher on the test, but according to P6, they were not interested in the test questions but in doing better at the game.

> *P6: I tried to become more accurate at killing the viruses when compared to the first time. It was more about the thrill of winning the game sooner, rather than the educational aspects. I had somewhat understood the basics covered in the first time the game was played.*

5.2.2 Retrospective Grades

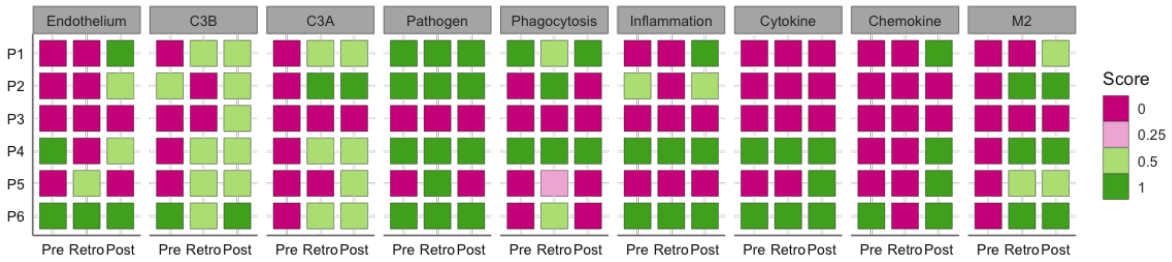


Figure 10: Comparing pretest, posttest, and retrospective score.

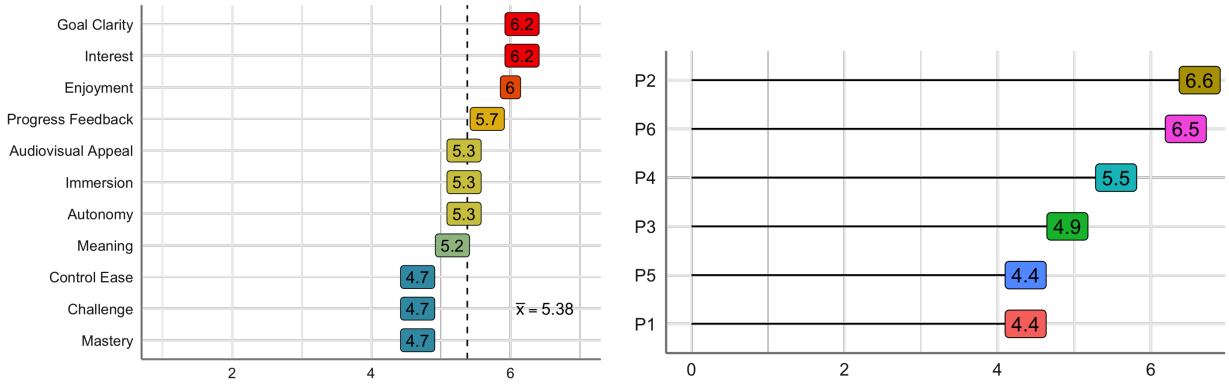
Figure 10 shows the results of grading the retrospectives (asking the participant "What happened in the game?" to elicit their recall of how the innate system fights an infection) and comparing those scores to each participant's pre and post scores. For elements like C3B, C3A, and the M2 Macrophage, we see that their gains in the posttest were also reflected in their retrospective (meaning that certain participants answered the pretest incorrectly,

but then included the element in their retrospective and scored higher in the posttest). Phagocytosis stands out as having multiple mentions in the retrospective but did not show improvement in the posttest results. Finally, several elements including Endothelium and Chemokine had increased posttest answers without mention in the retrospective. I have included the retrospectives and how I graded them in Appendix A.8.

5.3 Player Experience Results

5.3.1 Player Experience Inventory

Every question on the PXI uses a seven point likert scale from "Strongly Disagree" to "Strongly Agree" (I selected a subset of the full inventory and included them in the posttest; Appendix 7). The higher the measurement, the better the game performed for that measurement. Averaging across participants, the game scored 5.38 out of 7, which is good but leaves a lot of room for improvement (Figure 11a). Across measurements, players paired up into those that gave an average score of lower player experience performance (4.4), those that gave an average score for a higher player experience (≈ 6.5), and those that fell in the middle (≈ 5.2) (Figure 11b).



(a) Average PXI score per measurement.

(b) Average PXI score per player.

Figure 11: Average PXI scores.

I visualized the results of the inventory items into a divergent bar chart (Figure 12). According to the survey, the goals of the game were mostly clear, the game was enjoyable, the game roused curiosity in the participants, and most participants felt they had feedback about their progress. Mixed characteristics of the experience include ease of controls, audio-visual appeal, immersion, autonomy, and that the game felt meaningful. Finally, the weaker characteristics of the game are the facets of challenge and mastery.

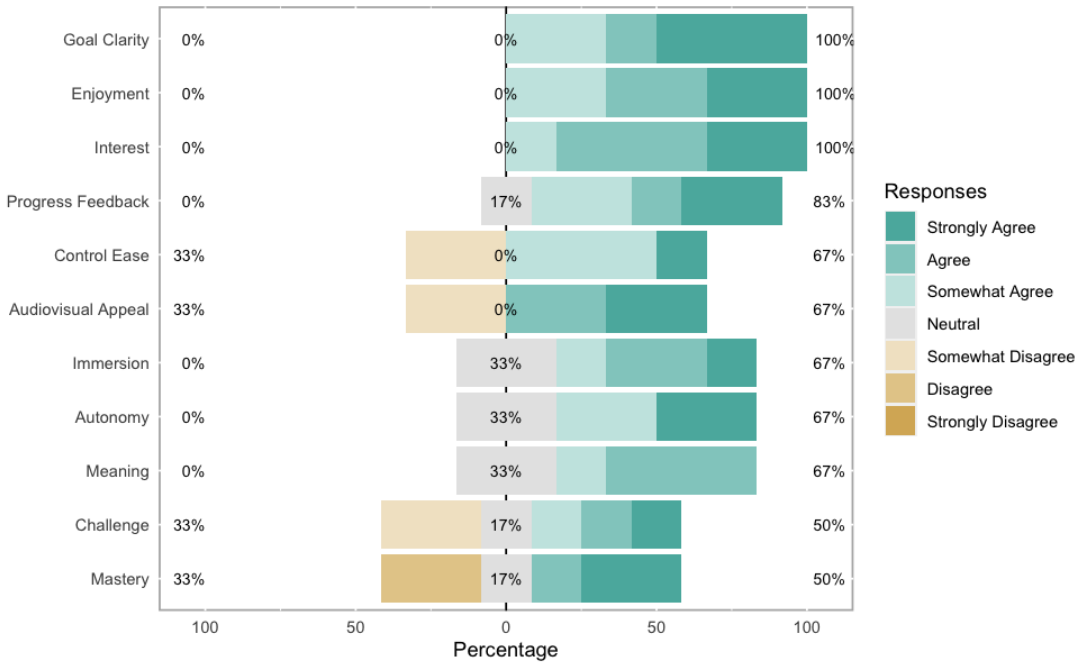


Figure 12: Player experience inventory results.

Using Spearman's correlation and a significance threshold of 0.2, I found strong positive correlations among certain PXI measurements (Figure 13). Both Challenge and Mastery strongly correlated with the most features including Control Ease, Immersion, Enjoyment, Meaning, Audiovisual Appeal, and each other. Where these two measurements fell on the scale, the rest seemed to follow. Figure 14 visualizes the gravitational-like force Mastery and Challenge have in the PXI scores. It is also intuitive to think the two are related since to feel a sense of mastery, the game should challenge the player but not be too difficult.

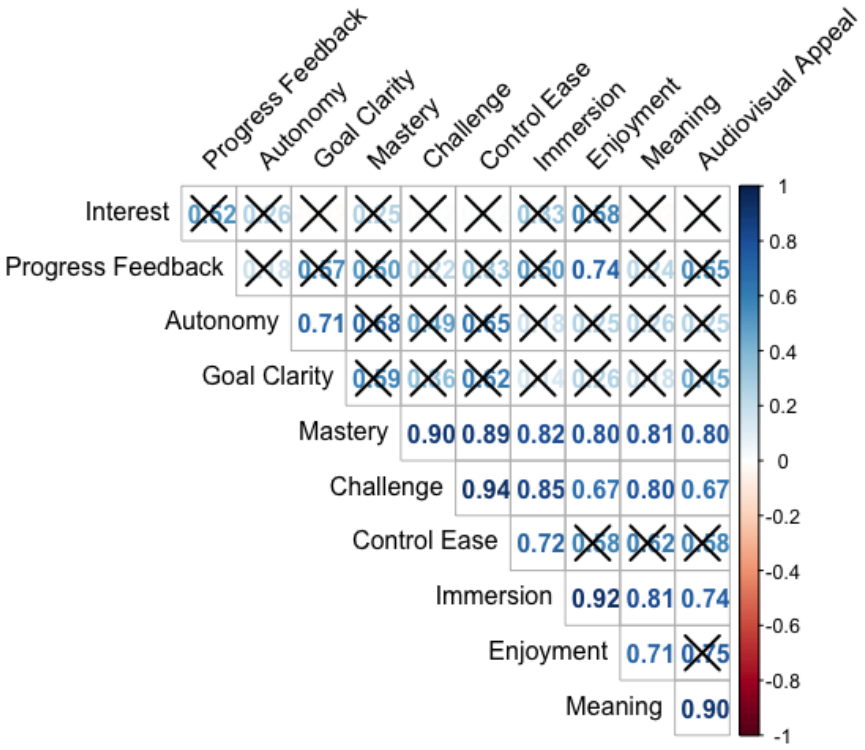


Figure 13: PXI measurement correlation.

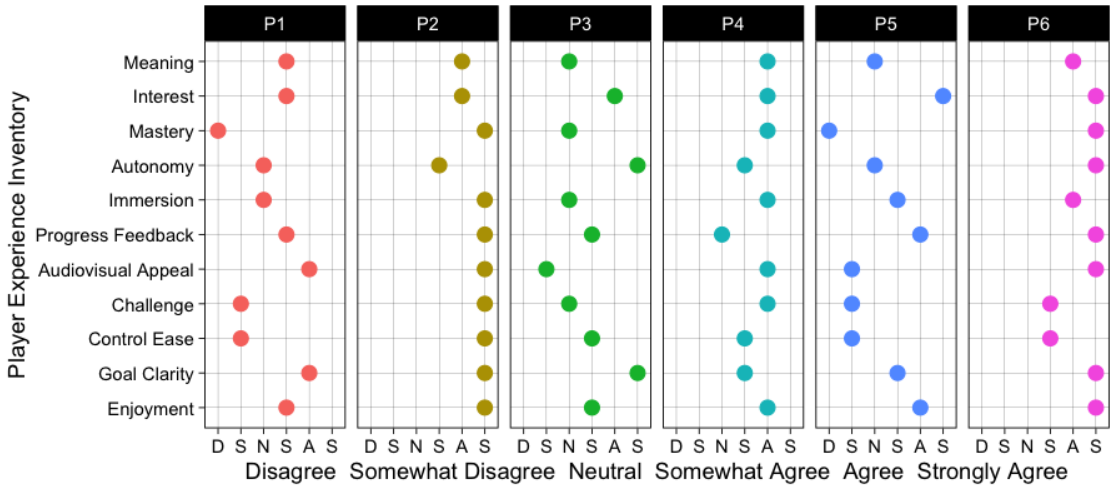


Figure 14: PXI results per playtester.

5.3.2 Player Performance

Player performance is the last facet of the player experience that I aimed to uncover in this study. This aspect of the player experience (how well does the player play the game), is evident in observing the player and can come up in the interview, however to quantify this measurement I needed a rubric. My lit review did not find studies that involved a rubric for player experience, so I borrowed from the world of sports and created a rubric that graded a player on their performance of nine skills in playing the game [10]. The skills were based on actions required to beat the game and one purely for measuring whether they won. The number of points awarded for each skill were based on their proficiency (Appendix A.7).

Figure 15 visualizes each participant's performance for each skill, which were colored as bronze, silver, and gold based on how many points I gave each. P5 was excluded from this part of the analysis as they were unable to play the game and share their screen due to graphical performance issues on their computer, so I was unable to observe most of their session and grade them. You can see that most participants excelled at collecting C3 and C3A, which was the easiest task in the game. Phagocytosis was the most challenging for participants, which was clear in observing many pathogens escaping the participants' grasp.

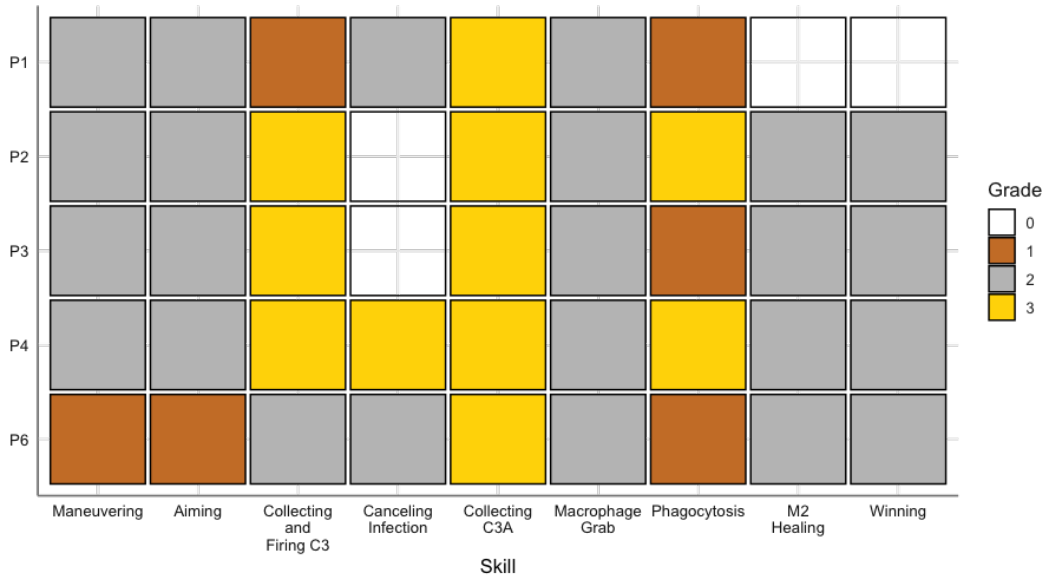


Figure 15: Player performance for each skill in gold, silver, and bronze.

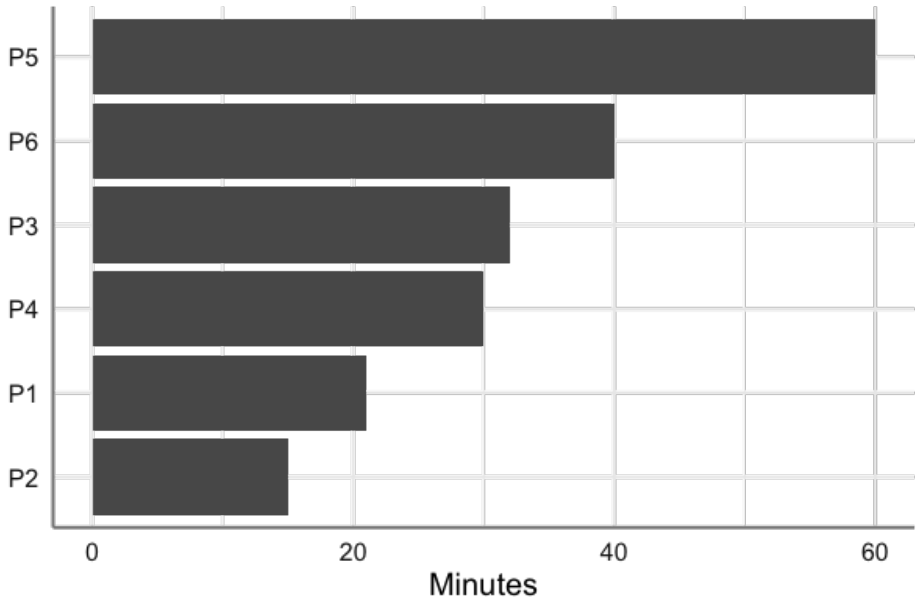


Figure 16: Minutes playing during playtest session.

This approach does not account for the level of difficulty each participant faced nor does it account for the length of time they played. I graded each participant on their last playthrough, and both P1 and P6 went beyond facing one live virus and played the harder levels that start with multiple live viruses. P1 also experienced much more aggressive viruses, possibly because of a bug where more graphics power speeds up the movement and A.I. of the viruses. Because of this issue, P1 was unable to beat the tutorial nor the other levels (and therefore did not get to use the M2 Macrophage). P2 and P3 also did not cancel an infection as they were able to defeat their one-on-one virus before it attempted to infect a cell.

Figure 16 shows the wide range of time each participant played the game during the playtest session. P2 requested for the playtest session to be no longer than one hour, so their in-game time ended up being the shortest. P5 had severe technical issues as the game was unplayable while sharing their screen, which led to multiple attempts until we decided to have them play without sharing their screen and relying only on their narration. While P6 experienced some bugs that increased playtime, they also played the levels the most number of times as they were determined to beat the live virus level outside the tutorial.

5.3.3 Thematic Analysis

RQ1: What is the player experience of Immuno-Defender, as determined by analyzing remote playtesting sessions and questionnaires?

My aim for qualitative analysis was to define the strengths and weaknesses of the experience. I also wanted to capture player suggestions and feedback that could improve the experience. Finally, I wanted to define what features were working towards the learning objectives and what features or issues got in the way. Evaluating the player experience involved combining and analyzing multiple layers of data. Video recording of the play session included the follow-up retrospective and interview questions and answers. I transcribed all recordings using Descript, and coded the transcriptions within Descript using Raycast[2] to insert codes using its snippet feature.

The codebook began with a priori codes and expanded with emergent codes that came from the transcripts, observations, and questionnaires. (Figure 17). Using Obsidian to perform deductive and inductive thematic analysis, I analyzed the resulting body of codes for patterns to describe the experience across participants [13]. During analysis, I reviewed each participant profile and pre/post test result to add context to the results.

Analyzing the codes across playtests led to more focused codes and eventually themes. Figure 18 is a visualization of the focused codes and themes within Obsidian. Figure 19 zooms closer to the codes associated with the themes about the games strengths (Table 3). Figure 20 zooms into the codes associated with the themes exploring the games weaknesses. Both sets of themes are listed in tables below along with selected excerpts from the transcripts (Table 3 and Table 4).

[2]Raycast is a free software that enables the user to execute various commands including creating a library of text snippets and inserting them into any program [50]

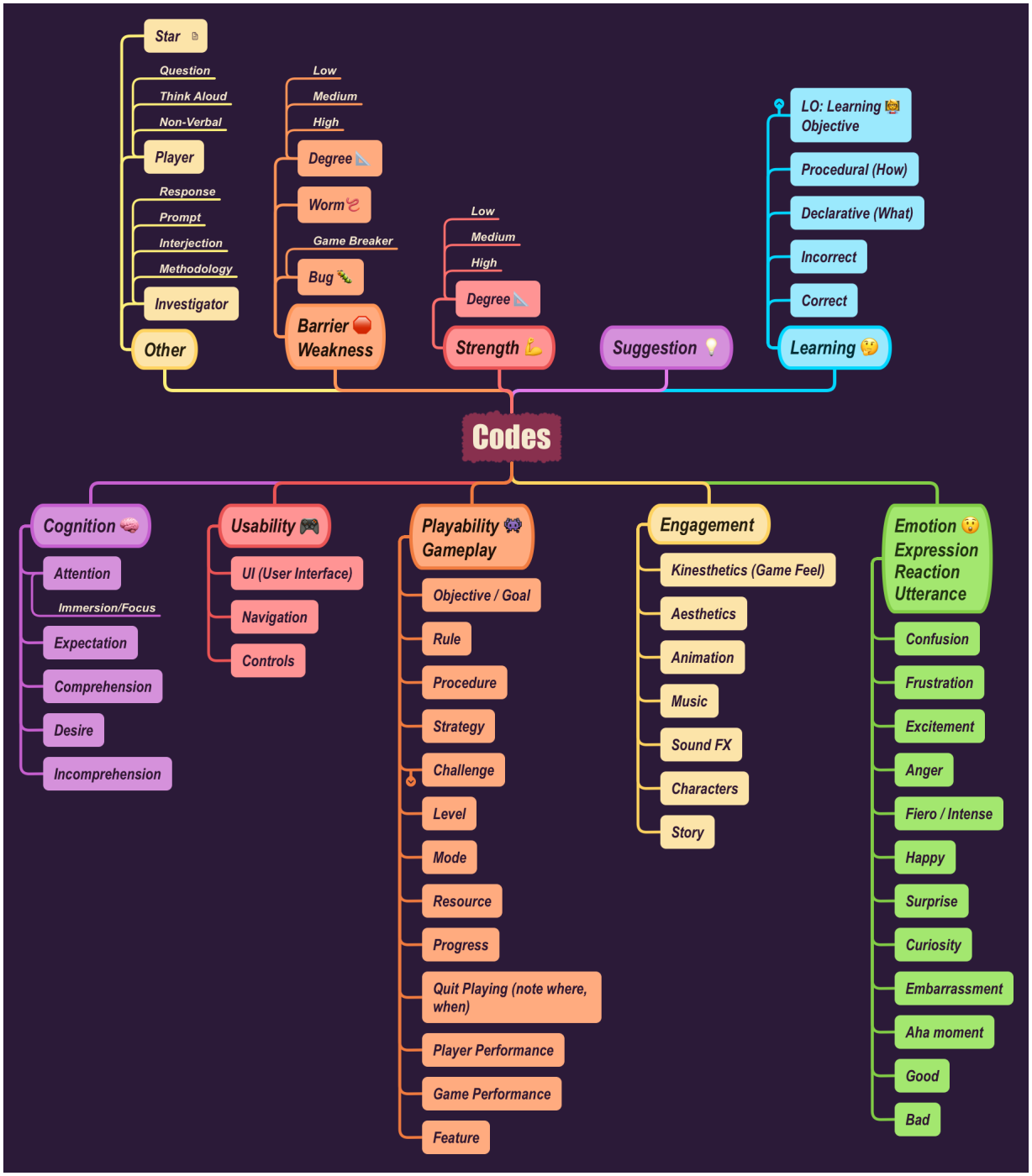


Figure 17: Coding Tree for Qualitative Analysis

Figure 18: Thematic analysis of codes.

Figure 19: Focused codes under the Strengths theme.

Table 3: Themes about the Game's Strengths

Theme	Select Excerpts
Alice (the A.I. Bot) improved the experience.	P2: "I think they did matter because I saved Alice. So that made me feel like this is what the immune system is doing for us. Like they're saving us."
NanoBot improved the experience and offered greater flexibility for game design.	P4: "We are kind of like letting the immune system get better... We're not really like taking over its responsibilities. I like that too."
Upgrade station has potential to be a knowledge check and participants like getting the answers right. Participants also liked the idea of gettings upgrades to the NanoDrone by completing Upgrade Station Challenges.	P2 nonverbally expressed being happy and excited getting the first answer right.
Visual presentation and animations were a highlight in the participants' experience. Moments including the inflammation groundswell and phagocytosis suprised participants.	P5: "I like the little animations." P6's favorite moments in the game were the macrophage eating the pathogen.
Participants felt a range of emotions including surprise, excitment, curiosity, humor, confidence, happy, and fiero.	P2: "I felt more confident and excited when I was fighting back, like when I was able to hit the virus and then get the macrophage, and then suddenly it was running away from me. I was like, I'll eat you..." P4: "I was feeling excited, and curious, and also happy that I'm kind of like learning about this."
The action gameloop was satisfying because it was not easy.	P2: "I liked the part where the virus started chasing me around and I had to fight back. I feel like that actually had stake. That was fun." P5: "Yes, I did it. Oh my god. Wow. This, oh wow. There's colors. Um, wow. That was, that's super satisfying."
Participants adopted immune system terminology when thinking out loud and answering interview questions.	P5: "Okay. And I'm going around, I'm picking up the compliment protein."
Participants had a positive impression of the experience.	P1: "I think it works well." P2: "I think it's like really neat and it's not overwhelming..." P4: "This is pretty cool." P5: "I thought it was good. I thought it was fun." P6: "It's a really nice game. I like it."
Most participants felt their decisions mattered.	P5: "I think that they did matter. I can see how your decisions in the game affect the outcome of the game."
Healing with the M2 Macrophage was satisfying to some but anticlimatic to others.	P5: "I like when you do things in a game and it affects the things around...that's probably my favorite part. Was the healing of the inflammation at the end." P4: "like right after the virus killing, it was a little bit anti-climactic for me, just like running around and healing things a little bit."
Players had "aha" moments of discovery, like finding out that live pathogens go after them and that C3B proteins can come off pathogens.	Observed for all players.

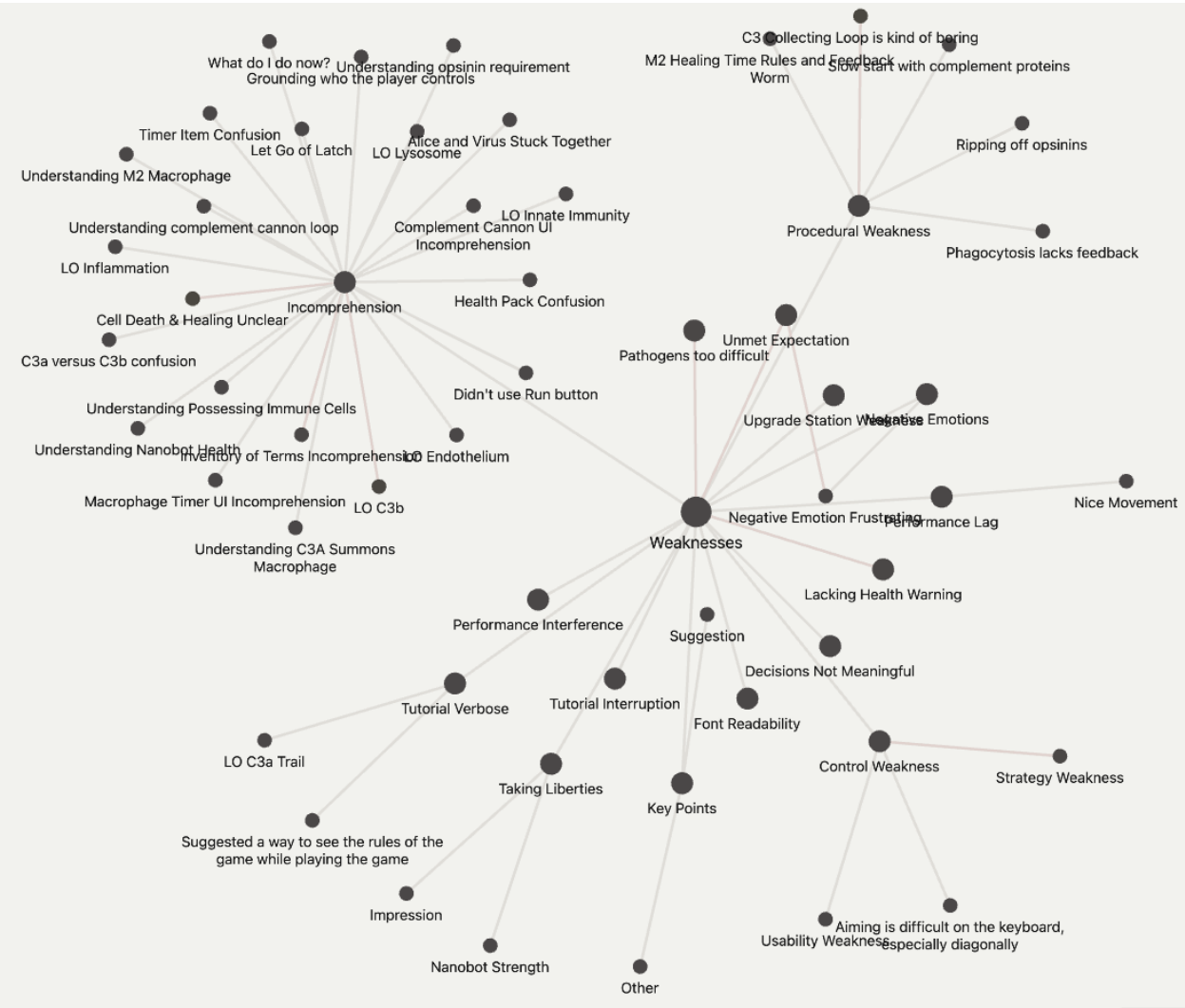


Figure 20: Focused codes under the Weaknesses theme.

Table 4: Themes about the Game's Weaknesses

Theme	Select Excerpts
Some participants skimmed parts of the tutorial and all would forget parts seconds later. Overall there was sentiment that the tutorial was too wordy.	P4: "I'm glancing over the text at this point. Kind of like skipping some parts." P5: "I already forgot all the instructions, but supposed to go collect things."
Participants with moderate graphics power had poor framerates, especially while sharing their screen. This greatly impacted aiming, movement, and invoked frustration in the participants.	P6: "Just lag a bit for some reason. I dunno. Cause zoom plus game maybe."
One participant with graphics power had pathogens that moved too fast to be eaten.	P1: "A little too smart. Yeah, it's way too fast."
Aiming was difficult for all players either due to performance causing lag or not being able to shoot diagonally despite pathogens being able to attack diagonally.	P2: "I couldn't aim diagonally, which was a bit hard because the virus was coming at me diagonally and I could only shoot like from up down left."
Participants were confused by the Term Index or did not use it.	P2: "Oh, are these ones that I haven't encountered yet?" P3: "Is this something you need to collect?"
Participants did not notice or fully comprehend important UI elements.	P2: "Is there a timer for the macrophage thing?" P5: "Wait, so I'm not allowed to get too close to it. Oh, okay. I have health. Do I?"
Participants were confused by the rules about the Timer Bonus Item and Healthpack Item.	P2: "I want to pick it up, but I don't know how." P4: "I cannot get this one though."
Some participants were unclear about what was happening during inflammation and cells dying from infection.	P2: "I was a bit like confused why the virus made a hole in here, and I think it's because like it reproduced."
Some participants were unclear about what they could heal and whether an area healed or not.	P2: "I am not sure if I can heal the hole. And if it's permanently like that." P4: "I see. So I did fix stuff. It was not very clear that I was fixing things."
Participants took some time to know that the macrophage could only grab onto C3B proteins.	P6: "I was really frustrated that, oh man, why is it not attaching?"
The macrophage mechanic has a major flaw where the player has nothing to do if none of the pathogens don't have C3Bs attached.	P6: "Okay. This is useless now. This is a useless macrophage."
Most participants did not use the "run" button.	Observed for most participants.
Participants that were defeated many times seemed to lose track of how much health they had and needed a warning.	P1: "Well, I'm bad at paying attention to my own health."

6.0 Discussion

6.1 PXI & Knowledge Acquisition

RQ4: To what degree does a participant's player experience, as captured quantitatively by their PXI answers, correlate to their score on a test that assesses their knowledge of immune system functions.

Figure 21 organizes the participants into three learning tiers, the participants that increased their score the least between the baseline and posttest (+15%), the players that increased their score by 25%, and the participants that saw the biggest jump in their score (30% and 45%). It then shows their baseline score to posttest score in the first column before moving through each PXI measurement with the y-axis representing "Strongly Agree" at the top level to "Disagree" at the bottom level.

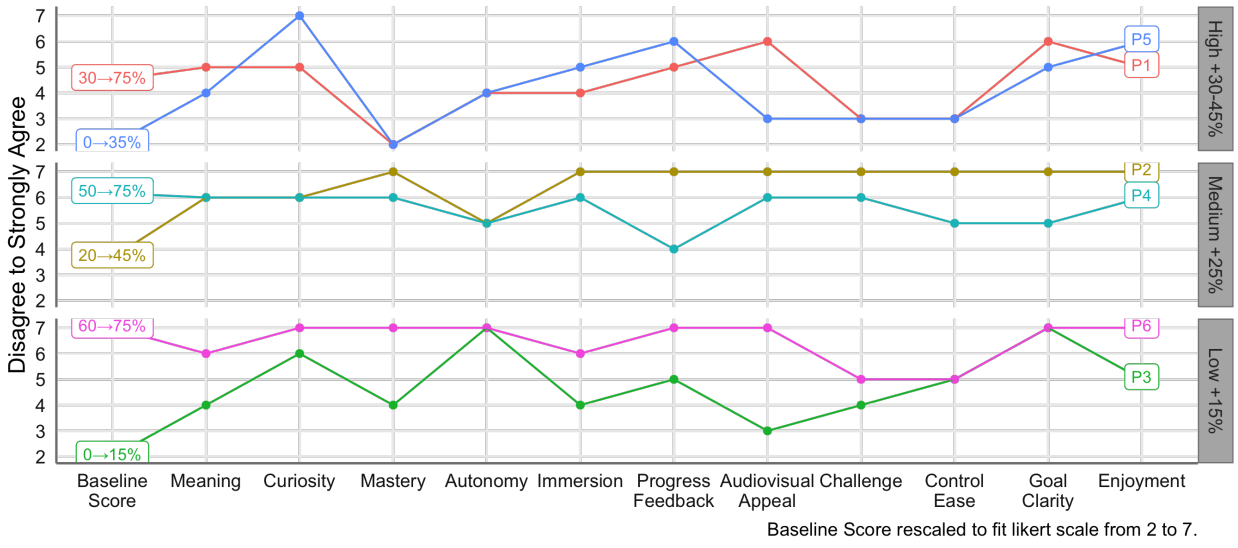


Figure 21: Comparing baseline score, score change, and PXI responses.

Notable insights concerning the top learners is that they gave the lowest response for mastery, control ease, and challenge ("Disagree"). Qualitative data backs their response

in that both participants struggled to beat the pathogens and struggled with controls. P1 struggled mainly because of a newfound bug where their computer's ample graphics power had increased the speed and force of pathogens that were impossible to catch and eat. P5 was on the opposite end of graphics power, which introduced several bugs including one that prevented the cannon from aiming reliably. Yet despite their struggles in playing the game, they managed to learn and increase their scores more than the other participants. However, due to their large increase in scores and overall lower player experience (averaging 4.4 as shown in Figure 11b) among a small sample, they highly contributed to the negative correlations in the Score Change column of Figure 22.

PXI	Baseline Score	Posttest Score	Score Change
Meaning	0.81	0.72	-0.09
Curiosity	0.02	-0.20	-0.31
Mastery	0.52	0.25	-0.58
Autonomy	0.24	-0.13	-0.97
Immersion	0.40	0.28	-0.13
Progress Feedback	0.04	-0.09	-0.13
Audiovisual Appeal	0.73	0.64	-0.06
Challenge	0.37	0.18	-0.44
Control Ease	0.17	-0.07	-0.63
Goal Clarity	0.05	-0.20	-0.64
Enjoyment	0.42	0.25	-0.18

Figure 22: PXI & Test Scores Coefficients

P3 and P5 had a baseline score of 0% and were "Neutral" in answering whether their decisions were meaningful. Others that started with a higher baseline were more agreeable to this measure.

P3 and P6 could not be more different in terms of baseline knowledge and PXI responses despite being in the same learning tier. P3 agreed less than P6 on most measures and started with a baseline score of 0%. P6 started with the highest baseline score (60%) and one of the highest PXI average of 6.5 (Figure 11b). Having such a high baseline at the starting line

limits how many learning objectives they would acquire to increase their score, unlike P3 who had plenty of room to grow.

Autonomy would not appear to be a factor in learning since the lowest learning group strongly agreed that the game gave them autonomy and the highest learning group were neutral.

Immersion likely suffered from the nature of participating in a remote playtest session since I asked participants to share their screen and talk out loud. It is hard to become immersed in something when not only are you being watched but you have to articulate your thoughts as well. Performance and bugs also impeded immersion. There was also division about the audiovisual appeal within the high and low learners, and overall participants either agree/strongly agree or somewhat disagree (no one being neutral).

6.2 Player Performance & Knowledge Acquisition

RQ5: To what degree does a participant's in-game performance of Immuno-Defender correlate to their score on a test that assesses their knowledge of immune system functions?

Figure 23: Player performance and test score change.

To answer RQ5, I first needed to determine if any of the records were outliers now that P5 was removed from this part of the analysis. Using Tukey's rule, I identified that P1's change in test score was an outlier [35]. Figure 23 visualizes their point on a chart that maps player performance and change in test score. Calculating the correlation coefficient with P1 gives a moderately negative relationship of -0.43 between player performance and score change (Figure 24). Removing P1 changes this relationship to 0.87, thereby emphasizing the issue with calculating such statistics with a small sample. Besides P1, we observe a positive correlation between player performance and change in score.

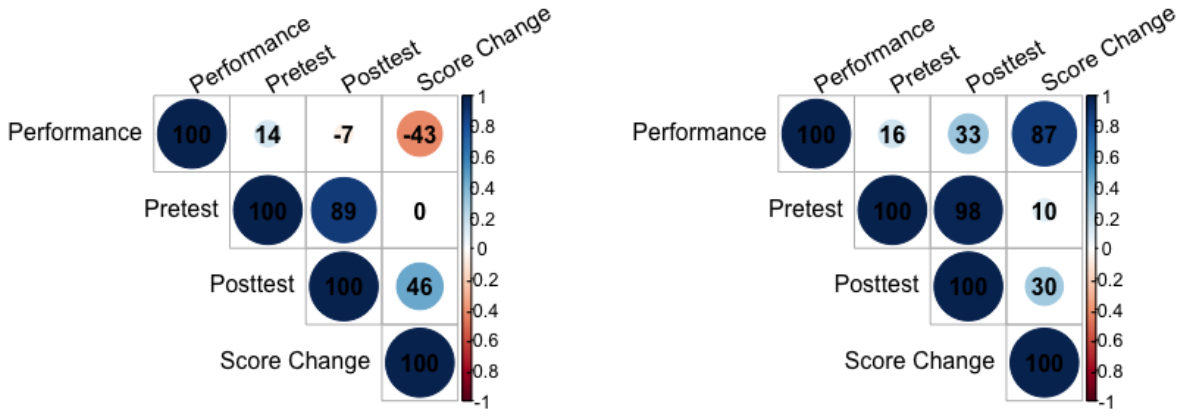


Figure 24: Correlation With and Without P1

While I made the rubric to address a research question, I also see it as blueprint to give players an in-game grade that encourages replay-ability and improving skills [9]. I would need to be mindful in what behaviors are rewarded points, since rewards can lead to behaviors that go against the intended experience [7]. For instance, P4 gained the highest performance score mainly by only facing one live virus and canceling an infection attempt (Figure 25). If a player was motivated to maximize their score by following this behavior, they would likely allow viruses to attempt infections just so that they can cancel them and increase their score.

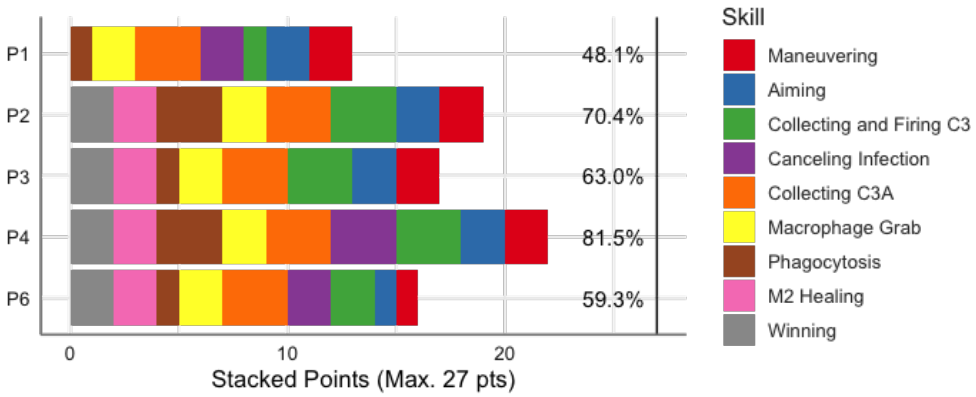


Figure 25: Stacked player performance points.

6.3 Evaluating Learning Features

The purpose of playtesting is to take a concept that has been engineered into a playable form and find out where expectations meet reality. The observations and feedback from the playtest can reveal the strengths and flaws in a game's design in the minds of players, and by analyzing the many layers of data we can find ways to improve the game's approach to teaching players through gameplay. Next, I will discuss a collection of features that I included in the game to aid knowledge acquisition, but were all clearly incomplete given the playtesting results. Then, we will examine the learning objectives, how they were featured in the game, and what the findings can tell us in order to improve their design.

Prototypes often include incomplete features for testing ideas before devoting resources to develop them further [52]. Normally a prototype tests one concept at a time, but this prototype includes a vertical slice of all aspects of the game since all parts contribute to the player experience and learning objectives. I added four features for knowledge acquisition and assessment: an in-game tutorial, a term index, an upgrade station, and debriefing.

6.3.1 The Tutorial

All games need a way to onboard new players to learn the game's objective, rules, and procedures. The simplest way of doing this is to provide a manual detailing the objective and rules of the game. A way more expensive yet engaging way of doing that is to include a tutorial level at the start of the game that guides the player in understanding how to play the game with low stakes. Following most modern games, I wanted the tutorial to be a level in the game that the player engages with like any other level. Expecting new players to not understand the innate immune system, the tutorial would have to do the heavy lifting without affordances that other games have. To make the tutorial more interesting, I included a character (Alice) to be the guide as well as a twist at the end of the tutorial. After the player completes all the steps in detecting an deactivated influenza virus, attracting a macrophage, phagocytosis, and healing, the game surprises the player with a live virus that they have to defeat and save Alice.

Figure 26: Alice the A.I. bot greets the player in the tutorial level.

Implementing an in-game tutorial doubled the scope of the project as it required a dialogue system, new uses of the camera system, and new game logic to control behaviors and events in the game to fit the flow of each lesson. The end result was a buggy tutorial that presented a new problem with each playtest, requiring the participant to restart the level if they hit a game-breaking bug. One particular game breaking bug occurred in the tutorial if the participant was standing right where the M2 Macrophage came up through the Endothelium and Alice (the A.I. bot) had not finished talking about the M2 Macrophage. Since the player possessed the M2 Macrophage before certain events dispatched, it would break the tutorial.

Due to this bug and others, some participants finished the tutorial and others had to abandon it and play a level that pit them against one pathogen to approximate the experience at the end of the level. If the participant missed any part of the tutorial due to a bug and was unable to replay that part, I would step in as Game Master and cover what the tutorial would have on its own. The tutorial's bugs ate up valuable time and prevented some participants from playing the game beyond the tutorial. Fortunately, the 1:1 battle with a pathogen covers the core of the gameplay loop so if the participant wasn't able to complete that part

of the tutorial, I had them experience it in a standalone level.

6.3.2 Term Index

Every term relating to the immune system is defined in an inventory that the player can access during gameplay. The implementation uses an inventory system and the terms appear as images in a grid with a description displayed on the left for the selected item (Figure 27). Only one participant (P6) explored the inventory during their playtest session, while others either ignored it or were confused by it (P2 and P3 both asked if the inventory was either logging what they have encountered or was a list of things they could collect in the game). While the feature was functional, it was also missing images for some terms, which adding to the confusion. P5 (the casual non-Action gamer that does not have visual/kinesthetic learning styles but instead prefers reading) wanted there to be incentive in the game to use the inventory but did not see the gameplay provide any.

> *P5: I think it would be cool if you were more incentivized to continue opening [the Term Index]. Cause that's like, I think if I would've opened that more, I would've gotten more knowledge ...about the actual immune parts.*

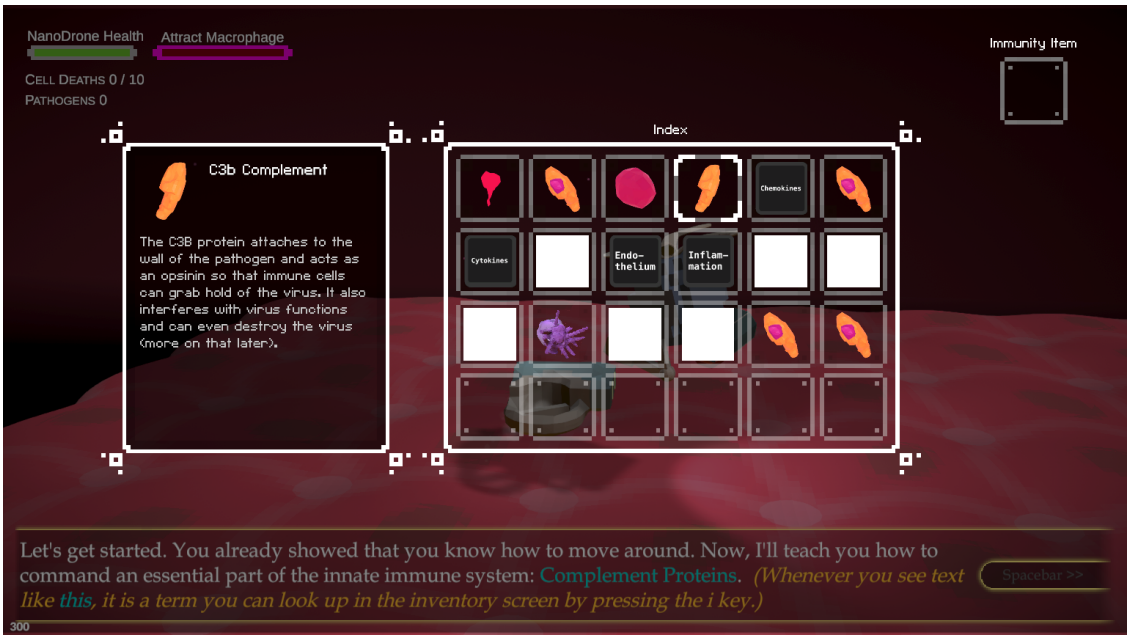


Figure 27: The Term Index provided descriptions for all immune system terms.

6.3.3 Upgrade Station

Anticipating that the player lacked incentive to review the Terms Index, I piloted a feature called the Upgrade Station after the playtest interview since the feature was separate from the core game loop. The Upgrade Station will provide temporary upgrades to the player if they can pass the "security check," which involves identifying parts of the immune system and their function (Figure 28). The prototype only included questions about C3B and C3A (questions that were unique to the pre/post tests) and were not functional in the build, so I had to moderate play as the Game Master. I discovered that participants liked the concept and wanted upgrades to make the game more interesting or easier. Some participants answered the questions correctly on the first try, while some needed more than one try. P2 (whom had their camera on) expressed being happy getting the questions right, lending that the feature also benefits the participant emotionally.

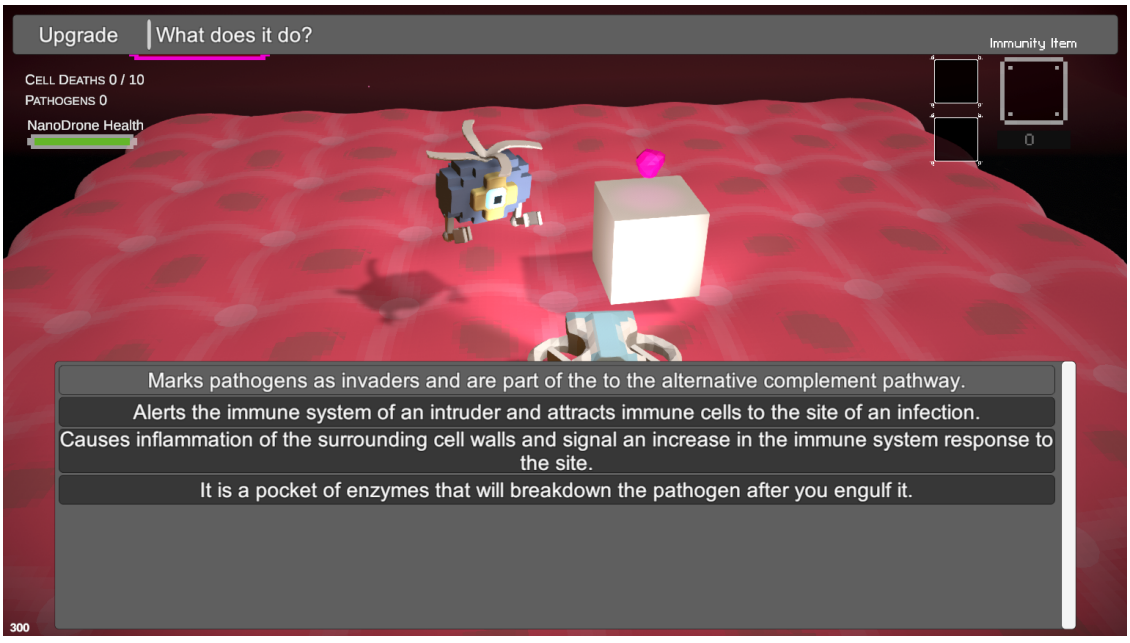


Figure 28: The Upgrade Station rewards correct answers with temporary upgrades.

6.3.4 Recap as Precursor to Debriefing

Debriefing with a learner can help facilitate comprehending new knowledge [29]. During my proposal, the committee suggested including debriefing in the game, and while it is a great suggestion I had no experience or knowledge myself of what debriefing entails as part of a learning project. While researching the topic, the scope of the game had also expanded with a full tutorial to ground the player in the rules and procedures of the game as well as the learning objectives. Already stretched thin, I programmed a simple recap at the end of the level to experiment with what a debrief could look like (Figure 29).

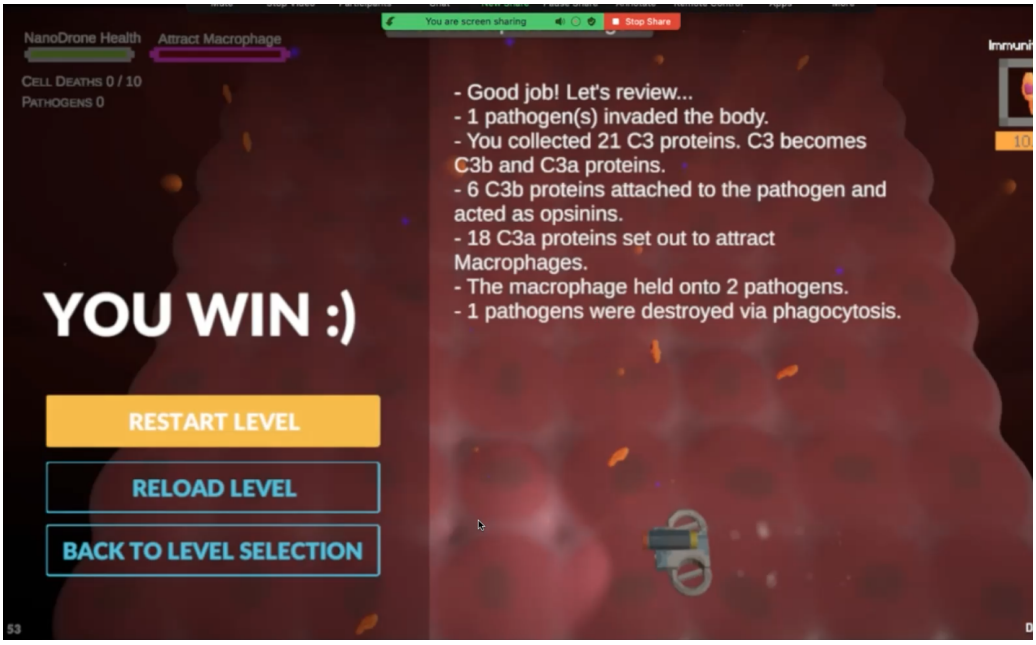


Figure 29: Players receive a recap of what happened when the game ends.

A proper debrief is more of a discussion and requires a facilitator to present to the learner the "what," "so what," and "now what" [21]. What happened? What is significant about it? And what's next for the learner to truly explore a knowledge domain? Doing this justice in a game is an entire project on its own, so a starting place is to at least provide feedback to the player with a recap of what happened. This is standard in some videogames to provide a recap at the end of a level to assess how well the player performed. For example, the game

Fortnite provides stats describing the player's combat proficiency and achievements at the end of a battle as well as during the battle using the Pause Screen [22]. Because I wanted this recap to become a debrief mechanism, instead of focusing on player performance I wrote the recap to be a descriptive list of what happened with specific counts based on the player's actions.

This feature is harder to evaluate as only two participants were able to test it due to programming bugs, and even those that did test it did not provide much feedback. When viewing their recap after finishing the level P4 commented, "Okay, nice." P6 played the game the most and so made the most use of the recap. After many defeats, they commented, "You collected 76 [C3 proteins]. What is the use of all that? 39 [C3b proteins]. Come on, man. Like the worst probably stats! Okay." P6 was getting frustrated that will all the resources they used to fight the pathogens as documented in Figure 30, they kept losing. But they kept retrying the level until they were victorious, at which point they made a "Woo!" sound upon viewing the recap. In the end, the recap is a good starting point in further developing a debrief feature, but more research and design in needed.

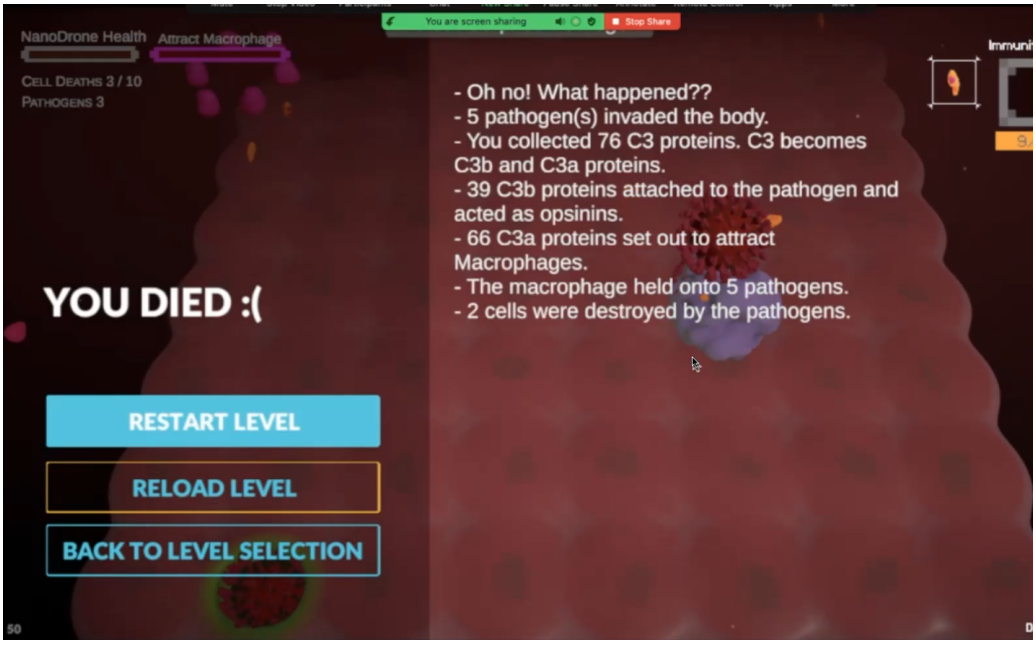


Figure 30: The quantity of complement proteins did not help P6 gain victory.

6.3.5 Floating In-Game Labels and Quest Text

In addition to the text-based approaches to teaching the player that I have covered (the level selection scene, Alice's dialogue, Term Index, Upgrade Station, and recap), I have also included text labels that display when quests are completed or objects appear. Some objects in the game would show a floating text label when they appear (C3, C3A, C3B, Macrophage, Cytokine, Pathogen, M2 Macrophage) and events like when the player completes phagocytosis for the first time display text in the middle of the screen (Figure 31). These are conventions in certain genres (strategy, action, RPG) and are a simple way to reinforce textual information.

Figure 31: Quest text in the middle of the screen and pop-up labels for C3A & C3B.

6.4 Evaluating Design Decisions & Learning Outcomes

6.4.1 Innate Immunity Featured in Level Selection

Test Question: The first line of defense against microorganisms that infect the body is referred to as ***innate*** *immunity.*

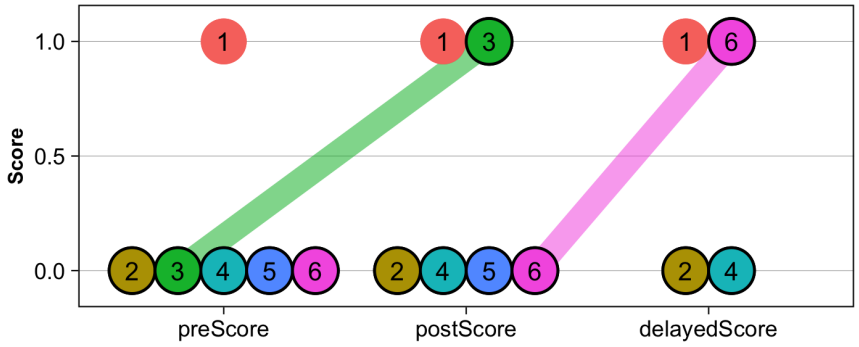


Figure 32: Innate test results.

Innate immunity is the all encompassing concept for the level and how it was taught to the player was through text in the level selection screen and a few mentions in the level. Figure 33 shows the part of the screen for the player to read a short line about the innate system. One participant answered this question correctly from the baseline and onward, but the outcome for the rest was that only one participant answered correctly in the posttest and one other answered correctly in the delayed posttest. Interestingly, the two participants that later answered correctly listed "reading" as one of their learning styles.

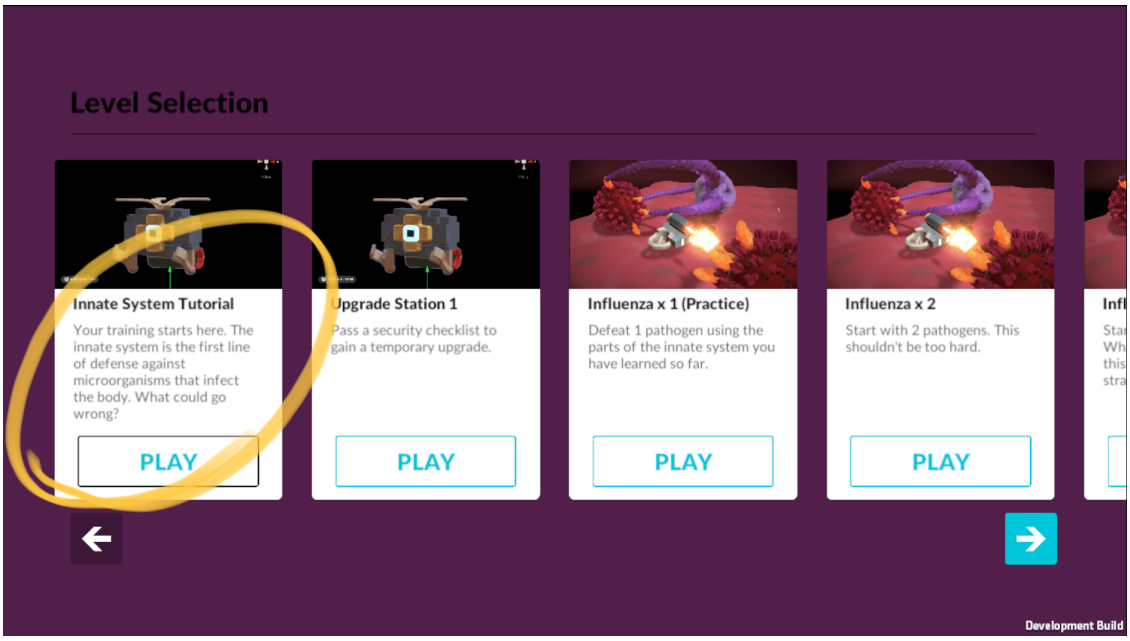


Figure 33: Innate System was featured in the Level Selector text.

I observed that all participants would pause to read the text (some paused longer than others), and yet the information did not stick for everyone. The design for this feature was overly simplistic compared to the innate system being anything but simple, and it did not include any explanation of what the innate system encompassed even though the items and characters the player becomes familiar with are an extension of that system. The book, *The Immune System*, introduces the innate system in contrast to the adaptive system [47]. A better approach for the game would be to include a pop-up text box with a detailed explanation of the innate system compared to the adaptive system before loading the level.

6.4.2 Endothelium Featured As the Level's Floor

Test Question: The thin layer of cells that makes up the interior lining of the blood vessels is called the ***endothelium****.*

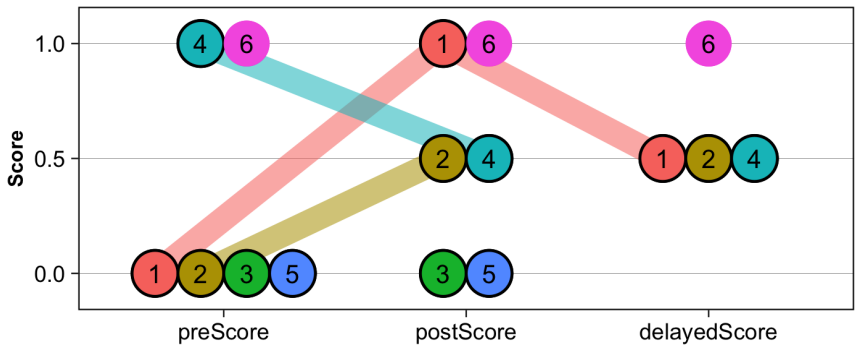


Figure 34: Endothelium test results.

Unlike the innate system learning objective, endothelium was featured as an object in the level as both the floor and as the host cells the player was tasked to defend. It was one of the earliest features developed for the game and changed through several iterations of internal testing. The endothelial floor is made of cells that can change shape and allow objects to push through during inflammation. The cells are also the target of the pathogen and can become infected and destroyed (and later healed by the player). The floor also acts as the level's boundary as the player is not allowed to move off the 8 x 8 grid of cells. The player learns what the floor is during the tutorial as Alice describes it, "These circles below are connected cells that make up the **endothelium**; the interior lining of the blood vessels. They form a permeable floor so that resources can move between layers in the body. We want to protect them from being damaged or infected by pathogens."

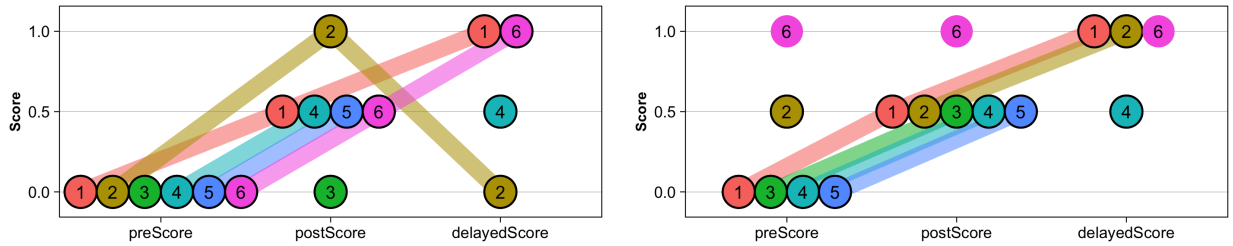
Figure 35: Objects like red blood cells traverse the permeable endothelium floor.

Three out of five participants ended up with partial points since they chose the similar sounding "epithelium"(P6 is excluded from the count as they knew the answer at baseline and onward). All participants seemed to interpret that the floor was made of cells judging by their answer to the tests or their retrospective (even P5 noted in their retrospective that they were on a "sheet of cells"). The main barrier is again remembering the specific term since so many participants were close to getting the question right, and being presented with two similar sounding terms on a test is likely to lead to some picking one and some picking the other. Outside of Alice's dialogue and the Term Index, there isn't any mention of the endothelium. This can be addressed in future iterations by referring to the cells in the user interface and recap as endothelial cells.

6.4.3 C3B and C3A Featured as Core Gameplay Items

C3A Question: What is the function of the C3A protein in the immune system response? What does it do?

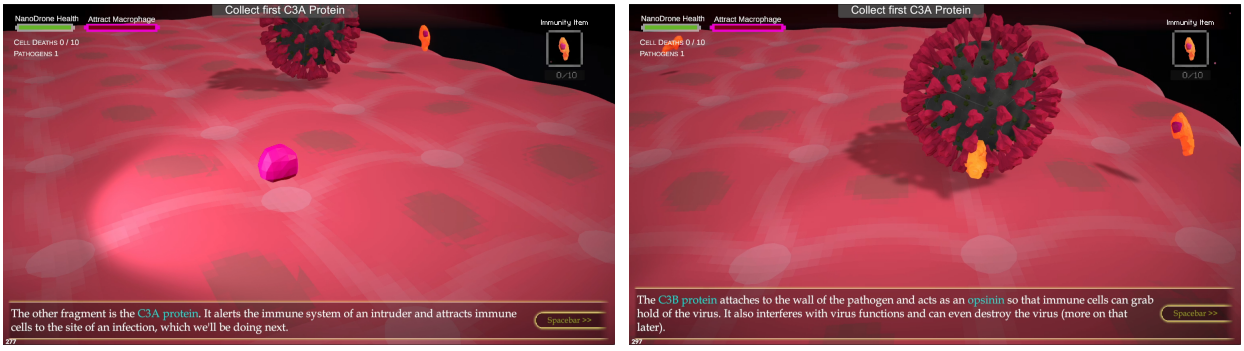
C3B Question: Which of the following complement components is an opsonin that binds to complement receptor 1 (CR1) on macrophages?



(a) C3A Protein (b) C3B Protein

Figure 36: C3A (open-ended) and C3B test results.

While the endothelium was important to the player's objective, it was not something the player directly controlled and required as much attention as the complement proteins. Complement proteins are a less known yet essential part of the innate immune system [1]. One particular complement protein, C3, floats around the human body until it comes into contact with a pathogen, whereby it marks it as a threat by attaching to receptors on the surface of the pathogen. Through a process called cleaving, it fragments into one part that stays attached and acts as an opsinin (C3B), and another part that floats around to attract immune cells (C3A).



(a) C3A Protein detached from C3 protein. (b) C3B protein attached to pathogen.

Figure 37: Complement proteins were gameplay essential yet new to most participants.

In the game, the player collects C3 proteins and launches them at pathogens whereby it cleaves and attaches a C3B protein to the pathogen and releases a C3A protein that falls to

the floor. The player then must collect enough C3A proteins to attract an immune cell (in this case a macrophage cell). In order to attract more macrophages, they must repeat the process of launching C3 proteins and collecting C3A proteins. The C3B acting as an opsinin is also necessary for the macrophage to grab onto a pathogen. If the player is a macrophage but the pathogens do not have any opsinins attached, then the player cannot grab them.

This core gameplay loop involving the complement proteins required the player to understand their function and process in order to defeat the pathogens. Since this demanded most of their attention and was critical to their success, they should have acquired the understanding of what these items were and their purpose and this should have been reflected in the test results. For the most part, this was the case. Everyone improved their score for C3B (excluding P6 who answered correctly at baseline) and all but 1 improved their score for C3A (P3 skipped all the open-ended questions in the pre and posttest, and notably stated that they were not interested in learning more about the immune system). The key issue for those that answered C3A and C3B partially correct is that they confused one with the other, or they described both in the C3A question. Table 5 lists the answers for each participant and many of the C3A open-ended answers described partially or completely the function of C3B, and those that answered the C3B question with "C3A" should have chosen "C3B."

Table 5: Test Answers for C3A and C3B.

	C3A (Open-Ended)			C3B (Multiple-Choice)		
	Pre	Post	Delayed	Pre	Post	Delayed
P1	Skipped	The C3A is the part of the C3 protein that binds to a pathogen, allowing a macrophage to grab onto it and ultimately consume it.	Unless I am confusing this with C3B, I believe the C3A is the protein that breaks off when C3 binds to a virus, such that C3A attracts a macrophage.	Skipped	C3A	C3B
P2	Skipped	Alerts the immune system of an intruder (pathogen) in the body	Skipped	C3A	C3A	C3B
P3	Skipped	Skipped	Missing	Skipped	C3A	Missing
P4	Skipped	It attaches to the viral pathogen's spike proteins and allows other cells in the immune systems such as the M1 macrophage to detect the virus and destroy it.	It attaches to the spike proteins of the viral pathogen so that the macrophages can recognize them.	Skipped	C3A	C3A
P5	Skipped	C3A (I think these were the orange things) bind to receptor sites on the pathogen that somehow strengthens the immune system and hurts the pathogen	Missing	Skipped	C3A	Missing
P6	learn the pattern build defence against pathogens	marks the pathogens and alerts marcophages	Signal the macrophages to be attracted to the virus	C3B	C3B	C3B

Reviewing the playtest sessions, retrospectives, and Upgrade Station trials, it was evident that some participants struggled to remember which letter went with which function while others did well:

- P1 got the two correct while thinking out loud in the playtest.
- P2 got the two correct in the retrospective and the Upgrade Station (although they referred to C3 proteins as C3Bs as well). In their pretest, P2 was asked, "Which questions did you find most difficult?" They answered, "The one asking about the function of C3A." They were then the only one that answered that question correctly in the posttest but left the delayed posttest blank.
- P4 stuck with calling the different proteins C3 despite reading out loud the dialogue.
- P5 struggled with names throughout the playtest and managed to identify the two objects on the second try during the Upgrade Station.
- P6 recognized each protein by name but would struggle with their functions during gameplay and the Upgrade Station, but correctly described C3B in the retrospective.

While designing the game I would also mix up the protein names myself. In fact, it is bad design to name things so similarly as it forces the player to work harder to remember the distinction. What the game needs is to teach players a mnemonic device to separate them (like *A* for "attract macrophages" and *B* for "binds to receptors").

6.4.4 Pathogen Featured as Adversary

Test Question: What is a pathogen and why is it a concern to the human body? (Open-ended)

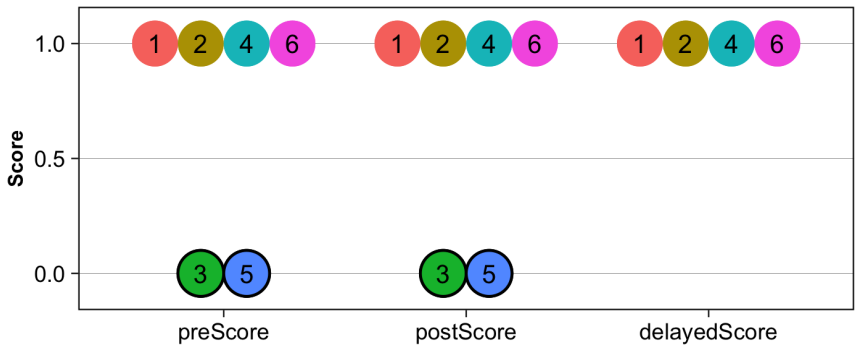


Figure 38: Pathogen (open-ended) test results.

Most participants were familiar with Pathogen except for two (one skipped the question in the pre and posttest, and the other skipped it in the pre and answered it incorrectly in the post). I would expect a higher chance of knowing or hearing about pathogens, especially given the increased discourse about viruses since the start of the pandemic. Looking at google search trends since 2004, Figure 39 shows that pathogen was the highest search term among five immune system terms included in the game (note as well the spike in the search cytokine at the start of the pandemic).

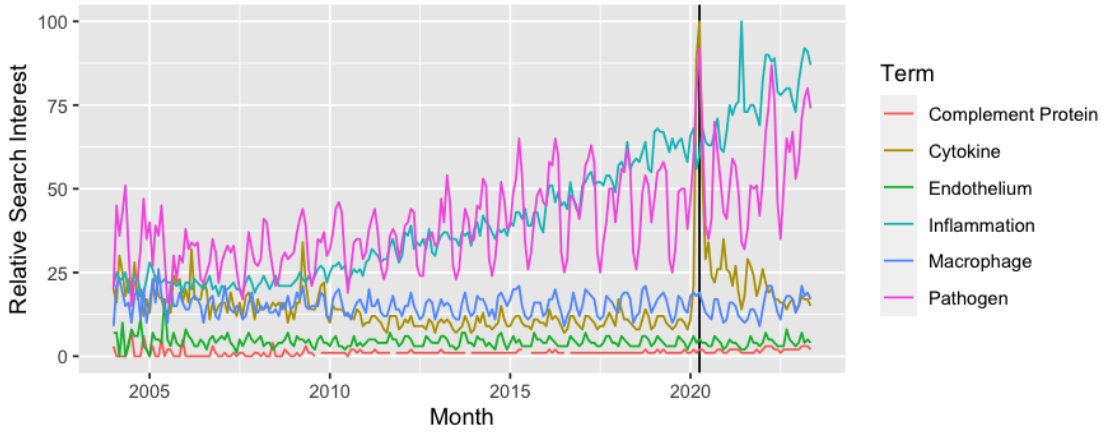


Figure 39: Immune system terms on Google Trends.

Pathogens were featured in the game as the player's adversary. The player's objective was to protect the endothelial cells and destroys the pathogens. It was clear from participants' behaviors and retrospectives that they did not struggle with this understanding. Perhaps if the question was not open-ended, the two participants would have answered the question in the pretest and likely answer it correctly. Open-ended style questions were difficult to four of the participants who said as much when answering the question "What questions did you find most difficult?"

6.4.5 Phagocytosis Was the Highlight Experience Missing from Outcomes

Test Question: Select the correct statement about innate and adaptive immunity. Correct Option: ***Phagocytosis*** *involves the uptake and killing of microbes.*

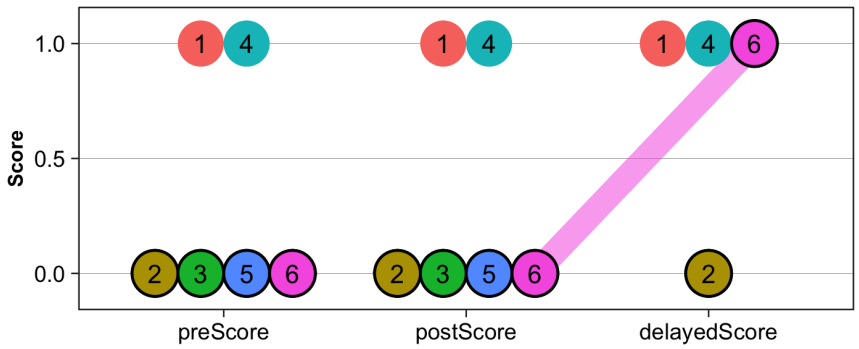
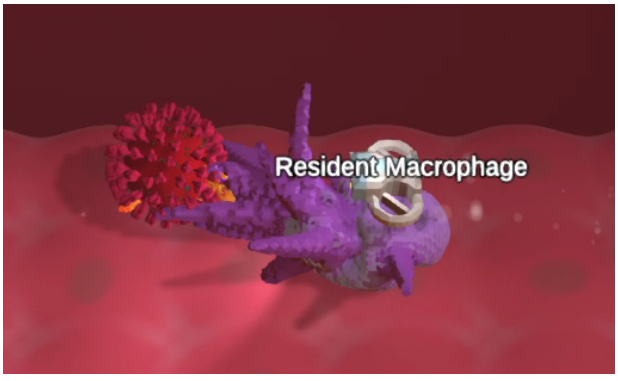


Figure 40: Phagocytosis test results.

Specific immune cells in the human body essentially eat pathogens in a process called phagocytosis, whereby they engulf the pathogen and dissolve it in enzymes [47]. This action was a focal point for the gameplay loop, which first casts the player as a vulnerable NanoDrone that can be destroyed by taking too much damage from collisions with the pathogens. After attracting and taking control of a macrophage (a type of immune cell), they turn the tables and can now attack the pathogens by grabbing onto them long enough to engulf them and complete phagocytosis (Figure 41a). The action is animated with tentacles stretching out from the amorphous cell to grab onto the C3B opsinins, the cell grows to engulf the pathogen, and then it is pulled in and devoured with a explosion of visual effects signifying the release of cytokine (Figure 41b).



(a) Macrophages grab onto the C3B opsinins.

(b) Macrophage pulling in pathogen.

Figure 41: Phagocytosis involves devouring pathogens and releasing cytokine.

This feature was a favorite among playtesters who felt fear as the vulnerable NanoDrone but then confident and powerful as the Macrophage. The aesthetics of the grabbing animation, eating animation, and release of cytokine was also satisfying and some participants said it was their favorite moment in the game or reacted while watching the animation. Participants also seem to understand the term and its meaning based on their retrospectives and when they would mention phagocytosis in the interviews.

What was your favorite moment in the game?

- *P4: I think the phagocytosis is nice.*
- *P6: Macrophage. That was very satisfying to watch. Like the entire animation of the virus getting engulfed.*

(Watching the phagocytosis animation during the tutorial.)

- *P2: Wow. I like the graphics. That's good.*

(During retrospective.)

- *P4: I can also control the macrophage when they arrive. And then I can kind of like phagocytose the virus by attaching to its C3 proteins. And then after that cytokines are released and then that increases inflammation.*

Despite participants qualitatively understanding phagocytosis, no one in the posttest answered the question intended to evaluate their understanding of phagocytosis correctly (besides those that did at baseline according to Figure 40). The reason I believe that Phagocytosis did not improve was because the question I chose from the Test Bank was not specifically about Phagocytosis (Figure 42). Participants that answered this question incorrectly mostly chose one of the first two options perhaps due to response order bias or the primacy effect (the question order was randomized but not the choices per question) [32][62]. Answering the question requires some understanding of adaptive immunity, which the prototype does not cover.

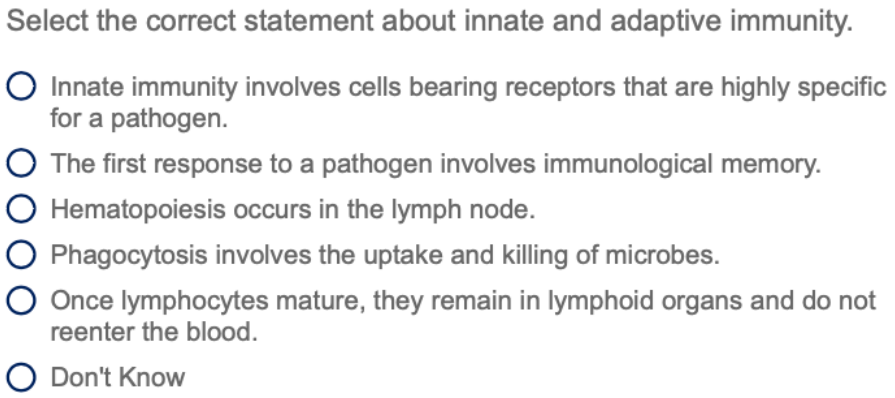


Figure 42: Question for Phagocytosis

6.4.6 Inflammation, Cytokine, and Chemokine Were Too Much Too Soon

Inflammation Question: What happens during ***inflammation****? (Open-Ended)*

Cytokines Question: When macrophages secrete ***cytokines****, an inflammatory response ensues*

Chemokines Question: The name given to cytokines that recruit cells to move toward areas of inflammation is ***chemokines****.*

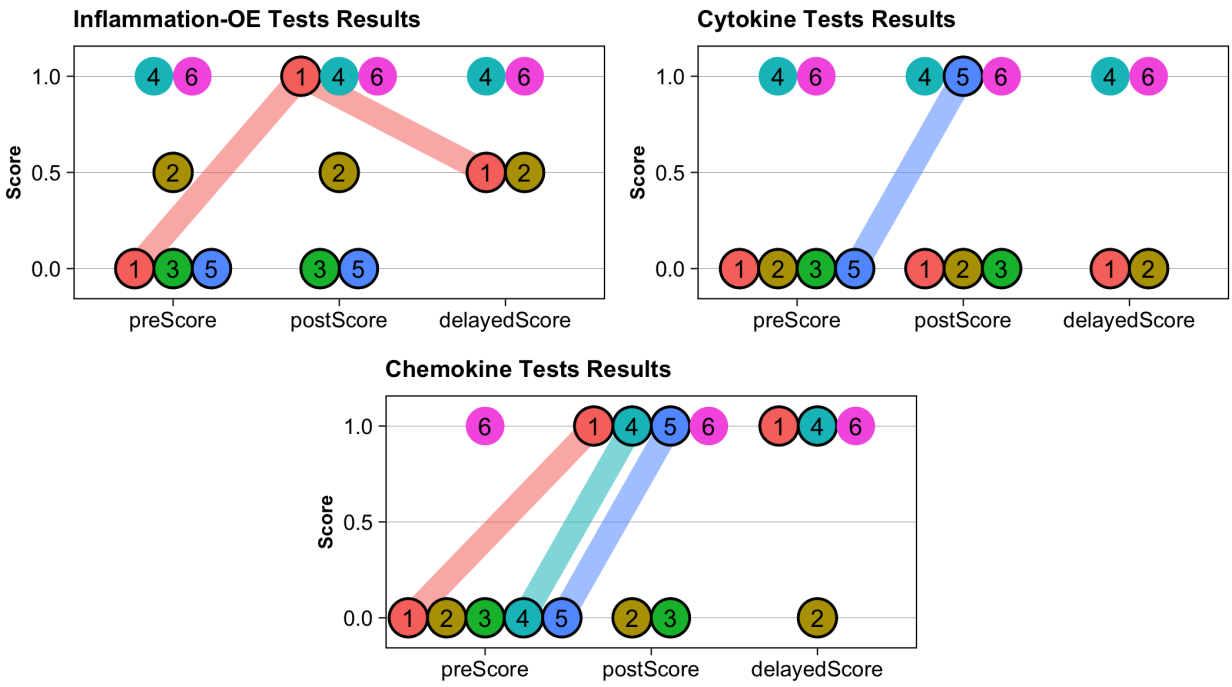


Figure 43: Test results for inflammation, cytokine, and chemokine.

A part of phagocytosis is the release of chemicals known as cytokines that trigger an inflammatory response. A type of cytokine is chemokine, which attracts more resources and immune cells to an infected site. Inflammation is a complicated topic in immunology and featuring it in the game was a challenging. After several iterations I ended up with a design where the macrophage releases particles that represent cytokine, which triggers the cells under the player to change shape and form pores in the endothelium so that resources could flow into the level. Then the player sees an upswell move from the top of the level to where they consumed the pathogen and a batch of objects are pushed up through the floor in a sort of geyser of particles, blood cells, and helpful items like a clock that extends how long they can be a macrophage, a healthpack to restore the NanoDrone's health, or an M2 Macrophage if they defeated all the pathogens.

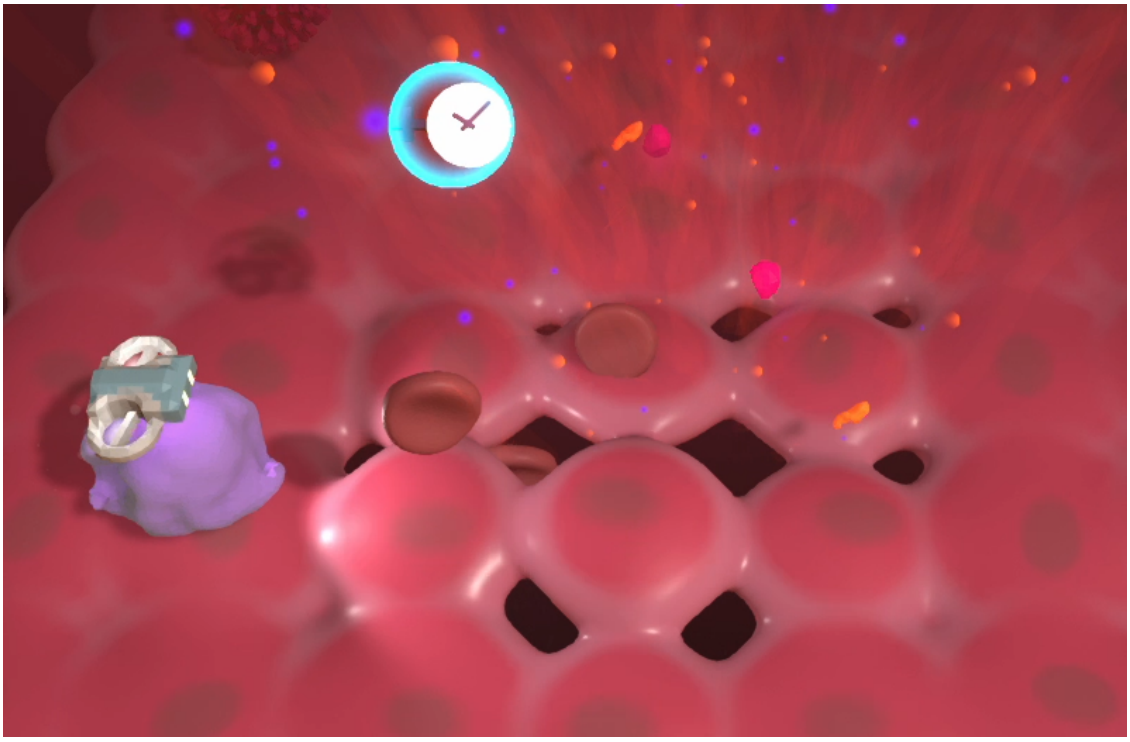


Figure 44: Resources flow through the porous floor during Inflammation.

Out of all the subjects in the game, inflammation was the most widely searched for in Google Trends and probably the term most participants at least heard about (Figure 45). Half of the participants received full or partial credit for the open-ended question for inflammation and one participant increased their score in the posttest (but later got partial points in the delayed posttest). Cytokine had some familiarity (as seen in the Google Trends spike at the start of the pandemic, Figure 39), but only one participant increased their score. Chemokine was the least known at baseline followed by a greater degree of uptake than the other terms in this section. Inflammation was rarely mentioned in the restrospective while cytokines and chemokines were not mentioned at all in the retrospectives for those that did not answer the test questions correctly at baseline.

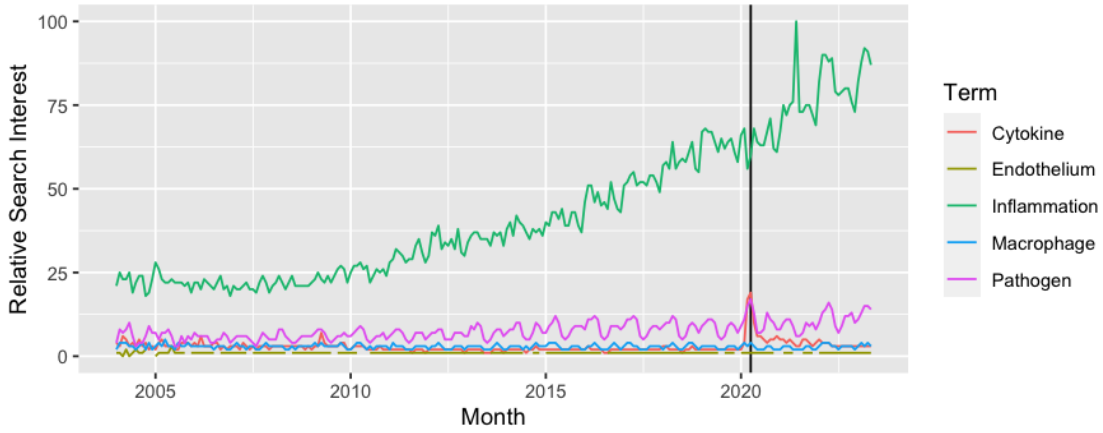


Figure 45: Google Trends including inflammation.

While inflammation was featured visually and functionally, it did not communicate clearly to the player all that was happening and perhaps that should be expected. While playtesting the game, the player is taking in a number of new concepts, symbols and meanings in order to learn the game and act as a player. Being human, players have a limit of how much information they can manage and even less that they will remember [59]. By this point in the game, the player has been learning a lot and likely maxed out their cognitive load:

1. How to control a NanoBot.
2. How to collect C3 proteins and aim and shoot them.
3. Knowing what are the pathogens that are trying to infect cells and replicate itself.
4. Knowing that they are vulnerable to taking damage so in order to defeat the pathogen they need to collect C3A proteins to attract a macrophage.
5. How to control the macrophage and grab onto C3B opsinins that came from the C3 proteins they attached to the pathogen.
6. How to hold onto the pathogen for three seconds to successfully complete phagocytosis.

Given this cognitive load, introducing something as complicated as inflammation was a bad design decision for the first level in a game, but I felt it was needed since it followed phagocytosis and was a key part of the innate immune system. A better approach would

be to save inflammation for the second level and build up the complete game loop more gradually so that the learning objectives had a slower pace [40]. This feature design also has some risk for misconception, as P2 for thought that the cells were being damaged when they changed shape to create pores. So separating inflammation for deeper playtesting is needed to avoid misconceptions.

It is odd that Chemokine had as much an uptake despite only being mentioned in Alice's dialogue and never mentioned in the interviews or retrospective. Since the Chemokine question was multiple choice, it is possible that some participants recognized the term from the game and simply chose it as a guess. Basing their decision on what they could easily recall (or in this case recognize), is known as the availability heuristic or availability bias [60]. The other options were caspase-recruitment domains (CARDs), inflammakines, adhesion molecules, and pyrogens. None of which were ever mentioned in the game so chemokines must have stood out.

6.4.7 M2 Macrophage Changed the Pacing for Better or Worse

*Test Question: What is the function of the **M2 Macrophage** in the immune system response? (Open-Ended)*

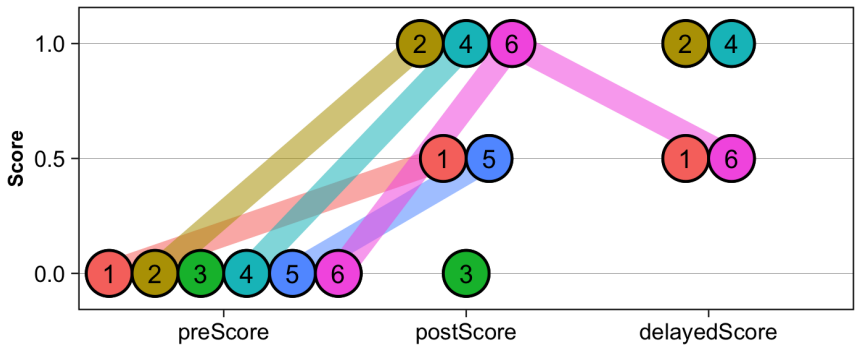


Figure 46: M2 Macrophage test results.

The M2 Macrophage was the final feature added to the game after learning about it from Dr. Empey. While the M1 Macrophage destroys pathogens and releases cytokines

to increase inflammation, the M2 Macrophage is a variation of the cell that actually heals cells and reduces inflammation [43]. This introduced an interesting change of pace in the gameplay to move from a Pac-Man cycle–evading and hunting–to a dénouement of the player taking control of an M2 macrophage and cruising about the level reducing inflamed sections and reviving dead cells (Figure 47).

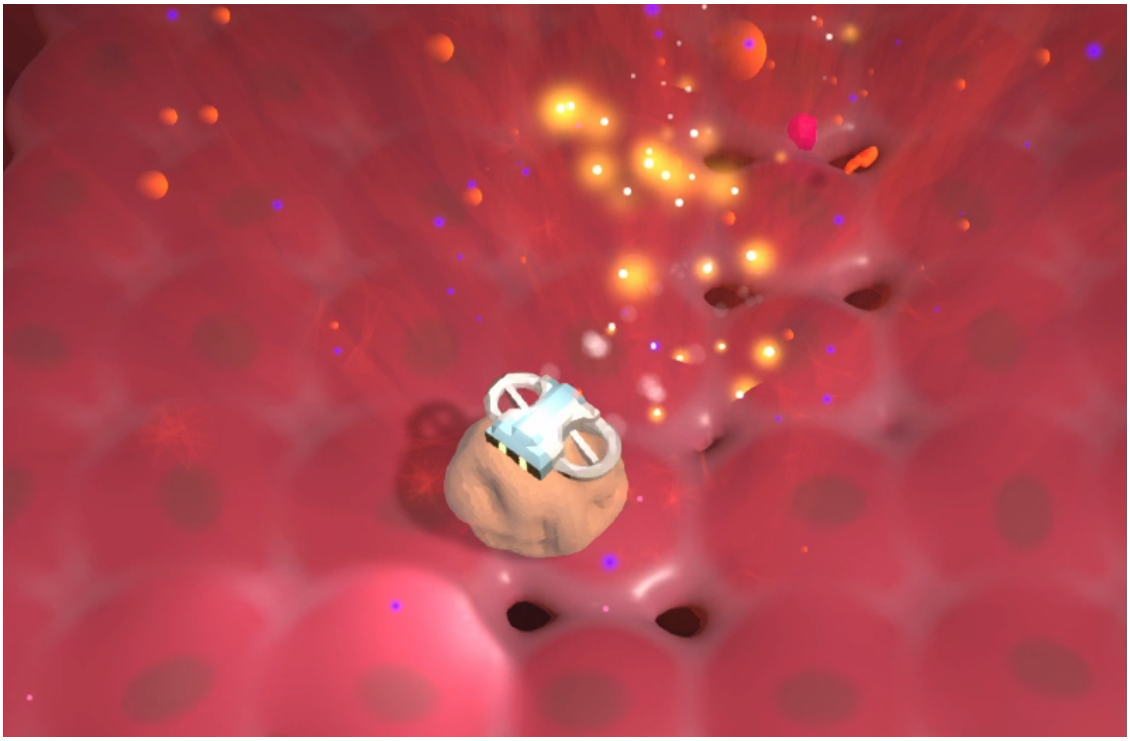


Figure 47: The player uses a M2 Macrophage to repair the site and end the level.

The immune cell was as new to me as it was to the participants as no one answered the open-ended question correctly at baseline, and all but two improved their score in the posttest. When the participant received partial points, it was because of a similar issue to C3A/C3B where they confused the M2 macrophage with the M1 macrophage. This kind of memory cross-wiring will continue to be an issue if the game continues to expand with more immune cells, pathogens, and environments as many terms can be similar (for instance Killer T Cell and Natural Killer Cell). I used different colors to visually identify the two

kinds of proteins and macrophages, but this does not help the player match a specific name to a specific object. P5 in particular would refer to objects by color in their retrospective because they couldn't remember the names. Further design research is needed to identify a better approach than relying on colors.

Table 6: Test Answers for M2 Macrophage Learning Assessments

	Pre	Post	Delayed
P1	Skipped	The macrophage envelopes pathogens and digests them in its lyosome (probably misspelled that) so that the pathogen cannot go on to harm the body or reproduce.	It basically consumes viruses, leaving them unable to infect healthy cells.
P2	Skipped	Repairs the cells damaged by the virus	It repairs the damages in the cell after the pathogen has attacked.
P3	Skipped	Skipped	Missing
P4	Phagocytosis of pathogens	It helps alleviate the infection symptoms such as inflammation in the surrounding region of the cell	It helps alleviate the inflammation response in the surrounding region of the infection.
P5	Skipped	I can't remember what that is. If it's the purple thing it kills the pathogen	Missing
P6	eats and digests out the pathogen cells	heal the damaged cells and cool the inflammation	To consume and digest the viral cell

P2, P4, and P6 all recalled the M2 macrophage for healing in their retrospective and completed the action in the game, while P1 was not able to finish the level and only read the tutorial portion about the function. (move this into above paragraph)

P4 felt that the healing portion of the level was anti-climactic and needed something as exciting as finishing the last pathogen. P5–who does not prefer action games–found it to be their favorite part. (expand on how the game would benefit from a greater variety of mechanics that are not all combat based since not everything about the immune system can be taught with combat, and plenty of games feature a variety of mechanics)

P5: I like the, I called it a trail of fairy magic... I like when you do things in a game and it affects like your surroundings or it affects the things around you. That's probably my favorite part, was the healing of the inflammation at the end."

6.5 Improving the Player & Learning Experience

6.5.1 Fighter Jet Versus Rambo

The controls were by far the biggest pain point for participants from the first to the last (Table 4). There were some performance issues that created a severe lag for some players, but overall players played the game like a 3rd-Person shooter and wanted to be able to move independently from aiming (also known as a "twin stick shooter"). The way the game was designed is that aiming follows the direction the player moves. This design decision stems from the first prototype of the game, which was a 3rd-Person drone simulator where controls were separated into moving in a direction and turning left and right. Pivoting the perspective to a top-down 3rd-person game, I thought that combining movement with aiming would simplify the controls.

This led to a classic mistake of game developers who play their game their own way until they find out that players have other expectations [42]. Usually this leads to games being more difficult to a new player than how the developer perceives the game to be. While this was also true for my game, it was the way I played the main character that was so different from the players. I had originally been playing the game using a gamepad and would fly around the level as if I was controlling a drone or fighter jet; rarely stopping and circling around pathogens like a bee while taking fly-by shots. Shooting forward made perfect sense.

The players on the other hand perceived the character not as a vehicle (like a fighter jet), but more like a gun-wielding mercenary that can move in any direction and aim independently of movement (like in games such as HELLDIVERS [5]). In the next iteration of the game, I plan to implement twin-stick controls to determine how it impacts the flow of the game. I can also search for top-down 3rd-Person games that successfully use movement and aiming as one.

6.5.2 All Out of Opsinins

Another glaring flaw in the game design is when a player is controlling a macrophage and has pulled off all the opsinins from the surrounding pathogens (Table 4, Figure 48). In

the current design there is nothing they can do except wait out the timer. This created a lull in the action and also is not entirely accurate. Macrophages can attach to pathogens without complement proteins acting as opsinins [26]. There are several solutions that could resolve this flaw and all should be tested with more focused playtests.

- Allow macrophages to grab onto pathogens without opsinins and have a lower success rate (thereby emphasizing that opsinins increase the macrophage's effectiveness. This would be more accurate and should be tested first.
- Rip off fewer opsinins if the pathogen escapes (this incentivices attaching more opsinins without leading to common scenarios of having nothing to grab).
- Give pathogens a stamina mechanic where each time they escape, they become weaker to escape again.

Figure 48: Opsinins break off as Pathogens escape the Macrophage's grasp.

6.5.3 Clearer U.I. & Reinforcing Terminology

One clear learning outcome is that while participants may learn the function of an object in the game, they may not learn the name. P5 in particular struggled with remembering the names of objects and events in the game. This can be difficult for action games that push players into the action part of the game and text is mostly ignored or is perceived as in the way. Moreover, players are notoriously averse to reading and it is common game design practice to avoid blocks of text [64]. This problem will only persist if the game were to continue with more levels exploring other parts and actors of the immune system. One solution would be to pivot the game into the Action-Adventure or Action RPG genre. Action Adventure games commonly involve stories that progress with dialogue and key events; both being opportunities to describe and reinforce learning objectives (such as storytelling scenes in any of *The Legend of Zelda* games [46] demonstrates in-game dialogue). Action RPGs and RPGs also involve story but go further by commonly having lots of UI elements and text since they involve more complex combat systems, such as the one seen in the *Pokémon* games [45].

6.5.4 Teaming Up with Immune Cells

One strength that emerged from the qualitative analysis was the use of the NanoBot and Alice the A.I.. The NanoBot provided a way for the player to embody a character in the level that would interact with immune cells and be vulnerable to the pathogens. Films and TV shows have used anthropomorphized versions of immune cells, which helps the viewer relate to the characters. *Osmosis Jones* teams up a white blood cell and a medication that are both portrayed as non-human characters but with human-like features [24]. Manga and TV Series, *Cells at Work!*, portrays all of the immune cell actors as humans fulfilling duties while pathogens are monsters [54]. My goal was to simulate the real appearance and functions of immune cells, but also have the player control a neutral character and interact with other characters to aid immersion and relatability. This led to the two nanotechnology characters.

Using this approach in an educational game comes with some risk of making an unintended argument that our immune system needs the help of technology to function. One

participant saw past the Sci-Fi elements but had to wonder:

> P1: I'm left thinking like, well, what does the body do without this little robot ? Like, my body doesn't have one of those little robots...

While my intention is to focus on the science of the immune system and not make arguments, I have to develop an awareness of my games procedural rhetoric [6]. I plan to add to my Playtest Guide a question that specifically explores the rhetoric of my game from the player's perspective (Appendix A.4).

7.0 Conclusion

Through a combination of qualitative and quantitative methods, I have evaluated the player experience and knowledge acquisition and retention of the prototype game, *Immuno-Defender*. Six students enrolled in the study and none had taken a college level immune system class nor played an immune system game.

By analyzing the changes in scores between pretests, posttests, and delayed posttests, I have determined that players gain some knowledge of the human immune system after playing the game and retain that knowledge after at least three weeks. However, the knowledge gained varied widely between players and qualitative data–including the retrospectives and interviews–revealed flaws in the test instruments (most of all the question for phagocytosis). The two methods also revealed a need for stronger presence and reinforcement of terminology, and that pivoting towards Action-Adventure or Action RPG genres with more story and text could help.

Qualitative findings and PXI scores gave an overall positive assessment of the player experience but also revealed its weaknesses. PXI scores were favorable for Enjoyment, Interest, and Goal Clarity, which aligned with observations and interview responses. PXI scores were less favorable for Control Ease, Challenge, and Mastery, which were also clear in how the game was too difficult for some and no one liked the controls and aiming. One disconnect between the PXI and the qualitative findings is that while during the interview and observations participants gave the impression they liked the visual style and animations, yet the average PXI score for Audiovisual Appeal was only 5.3/7 (Figure 11a). Finally, Immersion and Autonomy scored 5.3 as well and likely were affected by being observed, having to talk aloud thoughts, and being interrupted by bugs found in the prototype. Future research is needed in remote playtesting to determine the impact of these methods on PXI scores.

Observing the participants play sessions was critical in uncovering a flaw in the game design where the player has nothing to do except wait for the timer to run out if they ripped off all the opsinins while attempting phagocytosis. Among the many findings, this one is a direct issue with the game's design and how the feature works in the real world (in that

Macrophage's can attach to pathogens without opsinins). Further research and redesign is needed on this priority issue.

Comparing PXI scores and change in test scores required addressing one outlier that increased their score substantially but had a low PXI score. Including them results in a moderate negative correlation between PXI scores and change in test score, but excluding them resulted in a strong positive correlation. While I don't have statistical power behind this correlation or the t-tests I performed, I do have these instruments in place and can improve them before increasing the sample size.

In order to grade player performance in the game, I designed a player performance rubric and relied on the screen recordings for review. The results revealed what parts of the game were easier (collecting C3 and C3A), what parts were harder (phagocytosis), and what actions could be ignored (canceling infection sequences). I again analyzed the relationship between player performance and test score change by first identifying and removing an outlier (P1), resulting in a positive correlation. The next step is to build an automatic in-game system to grade player performance based on the rubric.

While the full set of methodology used in this remote playtest study has shown to uncover a wealth of insight into the prototype's player experience and learning outcomes, I discovered that it requires a great amount of time and effort to perform such a breadth of methods and analysis by a team of one and for a single iteration of development. Rather a more effective approach would be to perform more frequent smaller studies that focus on one or two features, instead of all the methods on a vertical slice of the game.

From this study I have determined that the game's design and development need a lot of work but have shown enough promise to merit further development. Visit ajmrkva.com/immunodefender to learn more about the project and its progress.

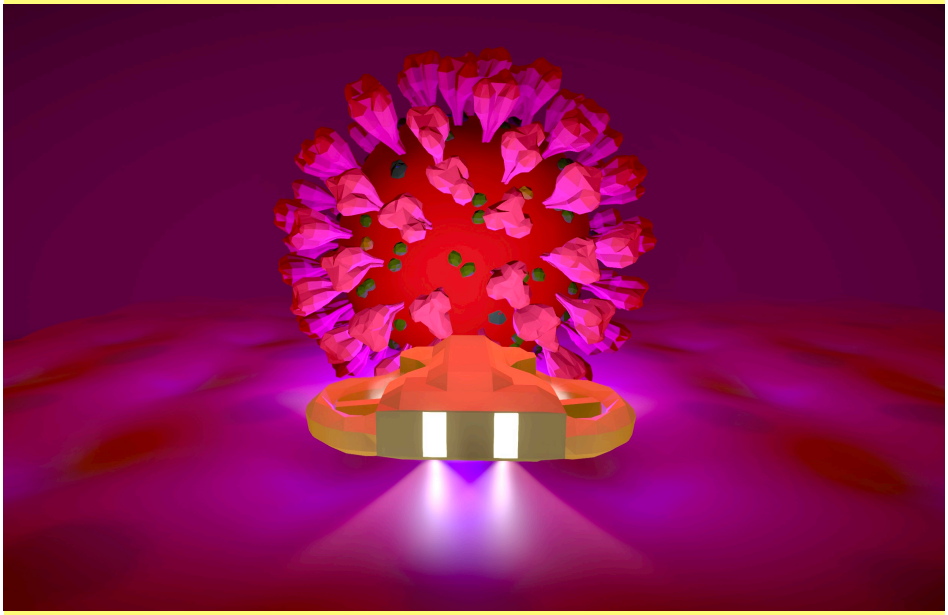
> *P6: If I were to like read this in a biology textbook, then I would never probably even remember the different complement proteins and how they function. But when you look at it in animation, it's very easy to understand, okay, C3A and C3B, like work together. And then these antigens and cytokines get generated after something gets destroyed and stuff like that. So, this is a really cool educational game.*

Appendix

A.1 Recruitment Flyer

Playtesters Wanted

Learn about the immune system while having fun



You are invited to participate in a research study and playtest *Immuno-Defender*, a 3D action game about the immune system.

The study requires that you have not taken college level classes about the immune system and are willing to complete a survey, pretest and posttest, a playtest interview, and a delayed posttest.

Please use the QR code or visit the link to fill out the recruitment survey and enroll.

Have questions?
Email Andy Mrkva at ajm240@pitt.edu

Visit this link to enroll!

tinyurl.com/playtest2023

A.2 Recruitment Survey

Introduction

Hello,

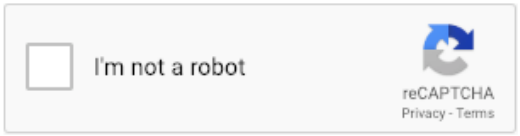
My name is Andy and welcome to my survey. I want to learn about your interest in games, the human immune system, and playtesting. Your input will help guide the design and development of *Immuno-Defender*, my game about the human immune defense system.

The survey should take approximately 15 minutes to complete, and all responses will be kept confidential.

Based on your answers, you will be invited to enroll in a research study where you will playtest *Immuno-Defender*. And if you are not eligible for the study, you can still enlist in being a playtester for the game.

Thank you for your time. I really appreciate it.

Please show this robot that
you are not a robot.



Immune System Knowledge

Before we get started, I just want to be clear that throughout this survey the term **immune system** refers to the **human immune system**.

With that out of the way, let's get started.

Are you a college student?

- ○ Yes
- ○ No

What is your degree program?

- ○ Undergraduate
- ○ Graduate
- ○ _____ Other (please specify)

What is your major?

_____

What styles of learning do you prefer? (Select all that apply)

- ☐ Visual (learning by visual representation like diagrams, charts, maps, images, animations, etc.)
- ☐ Auditory (learning by listening or speaking out loud like in lectures or reading out loud to oneself, etc.)

- [ ] Reading / Writing (learning by reading and writing)
- [ ] Kinesthetic / Hands-On / Experiential (learning by doing and experiencing through activities, role-playing, games, etc.)
- [ ] Social / Interpersonal (learning through social interaction, communication, or observation like with classmates or coworkers)
- [ ] Solitary / Intrapersonal (learning independently, alone, self-motivated)
- [ ] Other (please specify)

Please choose the level that describes your knowledge of the immune system.

- ○ Beginner: I know nothing about the immune system
- ○ Novice: I know a little about the immune system
- ○ Intermediate: I know what the average person knows
- ○ Advanced: I know more than the average person
- ○ Expert: I am an expert on the topic of the immune system

Are you interested in learning more about the immune system?

- ○ Yes
- ○ No

If yes, why are you interested in learning more about the immune system?

Have you ever taken classes on the subject of the immune system?

- ○ Yes
- ○ No

In what programs did you attend these immune system classes?

- ☐ Elementary School
- ☐ High School
- ☐ College, Undergraduate
- ☐ College, Graduate
- ☐ Professional (Not Academic)
- ☐ _____ Other

If you can recall, what were the names of these immune system classes?

Immune System Games

Please rate your level of interest in playing games about the immune system.

- ○ Not at all interested
- ○ Slightly interested
- ○ Moderately interested
- ○ Very interested
- ○ Extremely interested

Have you played games about the immune system? (Whether they be video games, board games, or card games.)

- ○ Yes
- ○ No

Please list the titles of the games if you can recall them.

What did you think about the game(s)?

Gaming Behavior & Technology

Do you play video games?

- ○ Yes
- ○ No

How would you describe yourself as a video gamer?

- ◯ Casual (play to pass the time)
- ◯ Beginner (recently started playing and value the time I play video games)
- ◯ Experienced (played for some time and have beaten one or more games)
- ◯ Expert (been playing for years and pride myself on my gaming skills and accomplishments)
- ◯ Professional (playing games is my job)
- ◯ _____ Other (please specify)

What is the average amount of time you play a video game in a given play session?

- ◯ Under 30 minutes
- ◯ Between 30 minutes and 1 hour
- ◯ More than 1 hour but less than 3 hours
- ◯ 3 hours or more

How often do you play video games?

- ◯ Daily
- ◯ Weekly
- ◯ Monthly
- ◯ Rarely
- ◯ _____ Other (please specify)

Please list the titles of video games you like to play most often (up to 5).

Please list the titles of your all-time favorite video games (up to 5). These might be the same as above but can include games you play rarely or haven't played in a long time.

What was the last game you bought? (Leave blank if you have not bought a game)

Thinking about a game you played recently, tell me what you **liked** about the game. Please include the name of the game and as much detail as you can about what you **liked** about it.

Thinking about a game you played recently, tell me what you **disliked** about the game. Please include the name of the game and as much detail as you can about what you **disliked** about it.

Do you subscribe to game libraries like Xbox Live, Playstation Plus, Nintendo Online, etc.?

- ○ Yes
- ○ No

How experienced are you with playing games that are…

	Not at all	Slightly	Moderately	Very	Extremely
2D Games	○	○	○	○	○
3D Games	○	○	○	○	○
VR (virtual reality)	○	○	○	○	○
AR (augmented reality)	○	○	○	○	○
XR (mixed reality)	○	○	○	○	○

How much do you enjoy playing games that are...

	Not at all	Slightly	Moderately	Highly	Extremely
Single player	○	○	○	○	○
Local Multiplayer (Couch Multiplayer)	○	○	○	○	○
Competitive Multiplayer (Versus others)	○	○	○	○	○
Cooperative Multiplayer (Teamed with others to achieve a goal)	○	○	○	○	○
Social Multiplayer (Sharing content or communicating with others)	○	○	○	○	○

How much do you enjoy playing games that are...

	Not at all	Slightly	Moderately	Highly	Extremely
Small groups (2-4 players)	○	○	○	○	○
Medium groups (5-12 players)	○	○	○	○	○
Large groups (more than 12 players)	○	○	○	○	○

What devices do you use to play video games? (Select all that apply)

- ☐ Nintendo Switch
- ☐ Playstation

- [ ] Xbox
- [ ] VR Headset
- [ ] Apple Computer (Mac)
- [ ] Apple iPhone
- [ ] Apple iPad
- [ ] Apple TV
- [ ] Windows PC
- [ ] Android Phone
- [ ] Android Tablet
- [ ] Google TV or Android TV
- [ ] _____ Other (please specify)

What input devices do you prefer to use when playing video games?

- [ ] Gaming controller (Xbox, Playstation, Switch, etc.)
- [ ] Touchscreen
- [ ] Keyboard and Mouse
- [ ] Joystick
- [ ] VR controllers
- [ ] _____ Other (please specify)

Game Genres

Think about the game genres you like most and select from the list below. Each group has sub-genres that I'll ask about next to fine-tune your selection to match your taste in games. If none appeal to you, please leave these blank and continue the survey.

- [ ] Action (Platformers, Shooters, Fighting, Steath, Survival, Rhythm, Arcade, etc)

- [ ] Action-Adventure (Non-linear, Linear, Survival Horror, etc.)
- [ ] Adventure (Text, Point-and-Click, Interactive Stories, etc.)
- [ ] Role-Playing (Action RPG, MMORPG, Roguelikes, Tactical, Sandbox, First-Person, Open-ended, etc.)
- [ ] Strategy (4X, MOBA, RTS, Tower Defense, etc)
- [ ] Puzzle (Logical, Trial-and-error, Hidden object, Tile-matching, Traditional, etc.)
- [ ] Simulation (Construction, Life, Vehicle, etc.)
- [ ] Sports (Racing, Baseball, Football, Basketball, Soccer, Boxing, etc.)

What action genres do you like?

- [ ] Platformers (Mario, Rayman, Legends, Sonic, etc.)
- [ ] Shooters (Halo, Call of Duty, Destiny, etc.)
- [ ] Fighting (Street Fighter, Mortal Combat, etc.)
- [ ] Beat 'em up/Hack and Slash (Streets of Rage, Ninja Gaiden, Devil May Cry, God of War, etc.)
- [ ] Stealth (Metal Gear, Sly Cooper, Splinter Cell, Dishonored, etc.)
- [ ] Survival (Don't Starve, This War of Mine, Minecraft, Fortnite, etc.)
- [ ] Rhythm (Dance Dance Revolution, Guitar Hero, PaRappa the Rapper, etc.)
- [ ] Arcade Style (Dig Dug, Pac-Man, Diner Dash)
- [ ] Action games in general
- [ ] _____ Other (please specify)

What action-adventure genres do you like?

- [ ] Non-linear (Legend of Zelda, Metroid, Castlevania, Ori and the Blind Forest, etc.)
- [ ] Linear (Tomb Raider, The Last of Us, etc.)
- [ ] Survival horror (Five Nights at Freddy's, Silent Hill, Resident Evil, Left 4 Dead, etc.)
- [ ] Action-adventure games in general

- [ ] _____ Other (please specify)

What adventure genres do you like?

- [ ] Text adventures (Rogue, Zork, etc.)
- [ ] Graphic or Point-and-Click adventures (The Secret of Monkey Island, Day of the Tentacle, etc.)
- [ ] Visual Novel/Walking Simulator/Interactive Stories (Gone Home, What Remains of Edith Finch, etc.)
- [ ] Adventure games in general
- [ ] _____ Other (please specify)

What puzzle genres do you like?

- [ ] Logical including Physics based or Coding (Portal, Braid, The Incredible Machine, Infinifactory, etc.)
- [ ] Trial-and-error / Exploration (Myst, Limbo, Monument Valley, The Room, etc.)
- [ ] Hidden object (Hidden Folks, etc.)
- [ ] Tile-matching (Bejeweled, Candy Crush Saga, Threes, etc.)
- [ ] Traditional (Word puzzles, number puzzles, Brain Training games, etc.)
- [ ] Puzzle games in general
- [ ] _____ Other (please specify)

What strategy genres do you like?

- [ ] 4X eXplore, eXpand, eXploit, eXterminate (Sid Meier's Civilization, Stellaris, etc.)
- [ ] Multiplayer Online Battle Arena / MOBA (Warcraft III, League of Legends, Dota 2, etc.)
- [ ] Real-time Strategy or Real-time Tactics / RTS / RTT (StarCraft, Warcraft, Warhammer: Dark Omen, etc.)

- [ ] Turn-based Strategy or Turn-based Tactics / TBS / TBT (Heroes of Might and Magic, X-COM, Wargroove, etc.)
- [ ] Tower Defense (Plants Vs. Zombies, They Are Billions, Sanctum 2, etc.)
- [ ] Strategy games in general
- [ ] Other (please specify)

What role-playing genres do you like?

- [ ] Action RPG (Dark Souls, Horizon Zero Dawn, etc.)
- [ ] MMORPG (World of Warcraft, Runescape, Ragnarok Online, etc.)
- [ ] Roguelikes (Spelunky, Rogue Legacy, FTL, etc.)
- [ ] Tactical RPG (Fire Emblem, Final Fantasy Tactics, etc.)
- [ ] Sandbox RPG (Assassin's Creed, Watchdogs, etc.)
- [ ] First-Person Party-Based RPG (Legend of Grimrock, Might and Magic, etc.)
- [ ] Open-ended RPG (Stardew Valley, etc.)
- [ ] Role-playing games in general
- [ ] Other (please specify)

What simulation genres do you like?

- [ ] Construction and Management (Football Simulator, SpaceChem, etc.)
- [ ] Life (The Sims, Dwarf Fortress, Animal Crossing, etc.)
- [ ] Vehicle (Truck Simulator, etc.)
- [ ] Simulation games in general
- [ ] Other (please specify)

What sports genres do you like?

- [ ] Racing (Need for Speed, Grand Tourismo, etc.)

- [ ] Baseball (MLB 20xx, etc.)
- [ ] Football (Madden 20xx, etc.)
- [ ] Basketball (NBA 20xx, etc.)
- [ ] Soccer (FIFA 20xx, etc.)
- [ ] Boxing (Punch-Out!, etc.)
- [ ] Sports games in general
- [ ] _____ Other (please specify)

What other genres do you like?

- [ ] Sandbox Other (Minecraft, Second Life, etc.)
- [ ] Casual Other (Peggle, Breakout, etc.)
- [ ] Party (Jackbox Party Pack, Warioware, etc.)
- [ ] Trivia (You Don't Know Jack, etc.)
- [ ] Fitness (Wii Sports, Wii Fitness, etc.)

Is there a genre you like but wasn't captured in any of the given options (including the "other (please specify)" entries)? If so, please describe the genre to the best of your ability and include a game title that fits that particular genre.

Otherwise, leave this blank and continue.

Other Games

Please list what board games and card games you like to play most often (up to

5).
Leave blank if you do not play board games or card games.

Designing Games for Players Like You

I want to design games for players like you, and the best way to do that is to share what I'm working on.

Are you interested in playtesting a game about the immune system?

- ○ Yes
- ○ No

What devices do you own or have access to? Select all that apply.

- ☐ Apple computer
- ☐ Windows computer
- ☐ Linux computer
- ☐ Chromebook
- ☐ Apple iPhone
- ☐ Apple iPad
- ☐ Android Phone
- ☐ Android Tablet
- ☐ None of the above

Good news, based on your answers you are eligible to participate in my study. Let's go over what your participation requires before you enroll.

The purpose of this research is to study the player experience and learning outcomes of Immuno-Defender, a 3D action game about the immune system. To participate, you will complete a pre-playtest questionnaire, then participate in an audio and video recorded Zoom meeting where you will playtest Immuno-Defender and answer questions. After the playtest, you are free to continue to play the game. Afterwards, you will complete a post-playtest questionnaire and then complete a delayed post-playtest questionnaire.

Risks of this study include the time commitment to participate in all parts of the study and the risk of a breach in confidentiality. To minimize the inconvenience of time commitment, you are free to complete each part of the study at your convenience. To minimize the risk of a breach in confidentiality, all identifiable information will be encrypted, data collected will be secured with password protected storage, and virtual meetings will be private. Potential benefits include learning about the immune system and/or being entertained by playing the game.

Your participation is voluntary, and you can withdraw from this study at any time by emailing the P.I. and request to be withdrawn from the study. If you choose not to participate, or if you do not complete the study, this will have no effect on your relationship with the University of Pittsburgh.

This study is being conducted by Andrew Mrkva, who can be reached at 412-224-0424, if you have any questions.

That was a lot of text. Please continue to the next screen.

Please enter your email address to enroll in the Immuno-Defender playtest

study. If you do not want to enroll, please continue to the next screen.

Please enter your email address if you would like to playtest *Immuno-Defender*. I will only use it to send invitations to playtest the game.

Please be patient, and I will contact you to playtest as soon as I can.

If you do not want to enroll, please continue onto the next screen.

Are you interested in hearing the results of my study or to be notified when the game is available to the public?

- ○ Yes, just the results of your study.
- ○ Yes, just the game being available to the public.
- ○ Yes, both.
- ○ No

If yes, please share your email below.

Demographics

What is your age?

What is your gender identity?

- ○ Female
- ○ Male
- ○ Non-binary or gender non-confirming
- ○ Transgender
- ○ Different identity (please specify)

Do you require any accommodations or precautions in playing digital games (e.g. avoiding flashing lights if you have photosensitive epilepsy)?

- ○ Yes
- ○ No

If yes, please describe any accommodations or precautions you need.

A.3 Pretest Questionnaire

Email Verification

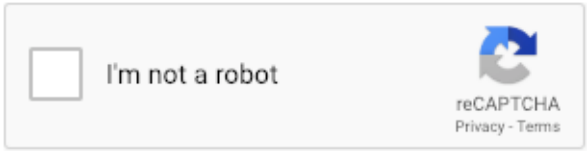
Thank you for participating in the Immuno-Defender pretest. Please answer the following questions to the best of your ability, and it is perfectly okay if you do not know the answer.

Please do not search for the answers online or elsewhere. The purpose of this pretest is to get a baseline of what you know before playing the game. And after you complete the pretest, still resist looking up any answers until you have completed your participation in the study (so at least until after you take the delayed posttest). Promise?

- ○ I promise I will not to look up answers until I have completed my participation in the study.

Please enter the email address where you received the link to this survey to ensure that we know who completed this test.

Please show this robot that
you are not a robot.



Immunity Quiz

Select the correct statement about innate and adaptive immunity.

- Innate immunity involves cells bearing receptors that are highly specific for a pathogen.
- The first response to a pathogen involves immunological memory.
- Hematopoiesis occurs in the lymph node.
- Phagocytosis involves the uptake and killing of microbes.
- Once lymphocytes mature, they remain in lymphoid organs and do not reenter the blood.
- Don't Know

The first line of defense against microorganisms that infect the body is referred to as _____ immunity.

- opportunistic
- innate
- adaptive
- primary
- central
- Don't Know

When macrophages secrete _____, an inflammatory response ensues.

- lysozyme
- defensins
- lymph
- sebum
- cytokines
- Don't Know

The thin layer of cells that makes up the interior lining of the blood vessels is called the _____.

- mucosa
- epithelium
- endothelium
- connective tissue
- lymphoid tissue
- Don't Know

Which of the following complement components is an opsonin that binds to complement receptor 1 (CR1) on macrophages.

- C3b
- C3a
- Bb
- Ba
- C3bBb

- Don't Know

The name given to cytokines that recruit cells to move toward areas of inflammation is _____.

- chemokines
- caspase-recruitment domains (CARDs)
- inflammakines
- adhesion molecules
- pyrogens
- Don’t Know

What is the name given to the intracellular vesicle in macrophages that have a concentration of degradative enzymes to completely degrade the pathogen?

- Opsonome
- Membrane-attack complex
- Lysosome
- Phagosome
- Phagolysosome
- Don’t Know

What is the function of the C3A protein in the immune system response? What does it do? (Leave blank if you cannot answer.)

What is the function of the M2 Macrophage in the immune system response? (Leave blank if you cannot answer.)

What happens during inflammation? (Leave blank if you cannot answer.)

What is a pathogen and why is it a concern to the human body? (Leave blank if you cannot answer.)

Review

How confident are you in your answers?

Which questions did you find most difficult?

Immuno-Defender Playtesting Guide

Andrew Mrkva

2023-02-04

Purpose

The purpose of this playtest is to assess the game's usability, engagement, how to improve the game, as well as the player's interpretation of the game, its process, and assessing their declarative knowledge and procedural knowledge acquisition through play.

The goal is to answer the following questions:

- Is the game comprehensible? (**Usability**)
  - Did the player understand what they were seeing and what they were doing?
- Is the game playable? (**Playability**)
  - Did the player understand the rules, the process, and attempt to beat it?
- Was the experience engaging? (**Engagement & Fun**)
  - Did the player express immersion or an emotional reaction to play?
  - Was the experience fun?
- Did they show they learned factual statements about the immune system from play? (**Declarative Knowledge**)
- Did they show they learned the process of how the innate system defends against pathogens (**Procedural Knowledge**)?
- What **changes** should be considered to improve the game and move the project forward?

Overall, analysis groups results into **barriers/weaknesses**, **strengths**, and **suggestions** for the game to be feasible and successful.

A Priori Codes

- **Usability**
  - User Interface (UI)
    - Navigation
  - Controls
    - Controls Comprehension
    - Controls Expectation

- **Playability / Gameplay**
  - Objective / Goal
  - Rule
  - Procedure
  - Challenge
    - Easy
    - Difficult
  - Level
  - Mode
  - Resource
  - Progress
  - Quit Playing (note where, when)

- **Engagement**
  - Kinesthetics (Game Feel)
  - Aesthetics
  - Animation
  - Music
  - Sound FX
  - Story

- **Cognition**
  - **Immersion**
    - Focus
  - Comprehension ->
  - Attention
  - Expectation

- **Emotion / Expression / Reaction / Utterance**
  - Confusion
  - Frustration
  - Excitement
  - Anger
  - Fiero / Intense
  - Happy
  - Surprise
  - Curiosity
  - Embarrassment
  - Aha moment

- **Learning**
  - Correct
  - Incorrect
  - Declarative (What)
  - Procedural (How)

- **Learning Objective**
  - Innate System
  - Endothelium
  - Complement Protein
    - C3
    - C3A
    - C3B
      - Opsonin
    - Macrophage
    - Phagocytosis
    - Lysosome
    - Antigen
    - Cytokine
    - Chemokine
    - Inflammation
    - M2 Macrophage
    - Pathogen
    - Endocytosis

- **Barrier/ Weakness🛑**
  - **Bug 🐛**
    - Game Breaker
  - **Worm🪱**
    - **Comprehension**
    - **Performance**
- **Strength💪**
- **Suggestion 💡**
- **Degree / Severity**
  - Low
  - Medium
  - High

Welcome (Icebreaker)

Thank you for volunteering to play the game.

Before we start, tell me about a game you played recently.
And what do you think about that game?
What do you look for in a game?

Let's Get Started

You will first play the game starting with the tutorial and can play the other levels afterwards. You can play for as long or as short as you like. It is up to you.

Please remember these 4 key things:

1. ***You are testing an early prototype**. The game is a work-in-progress and things are either not finished or untested, and we already know the game needs a lot of work. That's why we need you to help us understand what work is most needed.*
2. ***We are testing the game. We are NOT testing your skills and whatnot**. If you struggle in the game, it's most likely that the game is too hard and needs changing.*
3. *Try to **talk out loud your thoughts and actions** to help me understand you're thinking as you play. It may be hard to talk about what you are doing and thinking while you do it, and that's okay.*
4. *While I'm encouraging you to think out loud, **I will mostly be quietly observing and not say much** as I don't want to influence the experience. So if you don't hear me respond, I'm definitely paying attention, I'm just holding back my responses until after the playtest part of this.*

After you are done playing, I'll ask some questions and we'll have a discussion about the experience.

Think aloud / Talk aloud + Observation

During Play

Why did you make that choice?

Does that rule/control/action seem confusing?

What did you think that would do?

What is confusing you?

When they ask for help

Find out why

Ask them about their thoughts

Encourage them to try again

Assist last or when errors are hindering testing

Make note of expressions (verbal and non-verbal) to playing the game.

Retrospective / Skill Check[1] / Knowledge Check

This is actually the most important part of this playtest and the most experimental in terms of evaluating a learning game. We are going to either within the game or outside the game have the player explain what they do in the game, what is the procedure to playing the game, what are the parts of the procedure called, what do they do?

What happened in the game?

- What were you doing in the experience?
- What was the objective?
- What were the rules?

What did you do in the game?

- What were the procedures?
- What were the parts in the game?
- What were the items and the characters?
- What do they do?

What was your strategy?

What other strategies did you see or think of?

[1] Game User Research, Chapter 10, pg. 166

Playtest Questions

Overall, what did you think about the game?

What did you like most?

What was your favorite moment or aspect of what you just played?

What kind of emotions or feelings did you feel / notice as you played?
- What moments were the most fun?
- What was the most exciting moment of the game?

What did you not like about the game?

What was frustrating?

What moments felt like work, or were boring?

Was there anything more difficult than you expected in the game?

Was there anything unclear or confusing?
- What could have been explained better in the tutorial?
- What do you wish you knew when you first started playing?

Was there anything you wanted to do that you couldn’t?
- During the game, what action did you wish you could have done, but that was not part of the game?

Were your decisions meaningful, or did they feel like they didn't matter?

If you could change one thing, what would it be?

How would you describe this game to a friend?

Is there anything else that you would like to mention that we didn’t get to discuss?

Wrap-Up

Post-test prompt. Do it now or soon. You will get reminder emails to complete it.

Be on the lookout for a delayed post-test 3 weeks from the post-test.

Thank you for participating 😄

Post-Playtest Follow-Thru

Summarize this playtest and update the summary covering all playtests.

Compile Data into categories / sections.

Look for trends.

Make the compiled data readable.

Create Action Items.

For Issues, what was the cause and the impact? What is the consequence of not addressing the issue (severity)?

Include references, quotes, video clips supporting the findings.

List Interesting Callouts.

Keep track of changes made to the build.

Update Design Log.

Assess success of action items

For greater analysis, conduct keyword analysis or relationship mapping.

Interviewing Tips

Phrasing

Explain to me *how the dodge mechanic works?* Is better than *Do you understand the dodge mechanic?*

Example Questions

Example 1- The Puzzle
Bad: "Did you not understand the tree hint?"
Better: "Was there anything special about the tree puzzle?"
Best: "What could have helped you solve the tree puzzle?"

Example 2- A Trap
Bad: "You died a lot at the 3rd trap, what's up with that?"
Better: "Was the 3rd trap too hard?"
Best: "What did you think of the 3rd trap?"

Example 3- Powering Up
Bad: "What gave you speed boost?"
Better: "What did the red power-up do?"
Best: "What power-ups do you remember? What did they do?"

Don't Repeat Questions

Asking a question will influence the player's thinking. So be thoughtful when to ask a question and don't expect to have another chance.

Repeat Back Answers

In your own words, repeat back their answer from time to time.

Answering Questions

If the player asks something, try to get at their understanding or expectations.
- *'What would you expect?'*
- *'How would you expect it to work?'.*

If a player wants to know something and you don't want to influence them:
- *Unfortunately, I'm not going to be very helpful right now, so I will leave that with you. If it's still unclear at the end of the session, we can talk about it then*

Avoid Leading Questions and Phrasing

- *BAD: What made you think that?*
- *GOOD: How do you know that?*

Get to the Root of the Problem

- *Why is that?*
- *What caused that?*
- *How did that effect you playing the game?*

Glossary

Bug🐛: In-game unintended technical issues.

Cause: What about the game made the event/issue occur? (Related to Impact)

Feature is a function or asset in the game. Basically, anything that was put into the game or how the game works is a feature.

Game Breaker: Bug or gameplay element that breaks the game or blocks the player from continuing the game.

Impact: What is the impact of the event/issue? Or what are the consequences of not addressing it (how severe)?

Level of a game.

Mode of a game.

Procedures: Method of play / Actions a player may take.

Resources: Assets used in pursuit of the objective must have utility and must be scarce.

Rules restrict actions the player may take and trigger events (if x then y happens).

Worm🪱: Design issues in the game that in most cases was intended but playtesting shows wasn't a good idea.

Sources

Bracke, C. (2020, May 13). *What To Do After The Playtest: 20 Playtesting Questions That Set Players Up To Give Great Answers - Entro Games by Chris Backe*. Entro Games. https://www.entrogames.com/2020/05/what-to-do-after-the-playtest-20-playtesting-questions-that-set-players-up-to-give-great-answers/

Drachen, A., Mirza-Babaei, P., & Nacke, L. (Eds.). (2018). *Games User Research*. Oxford University Press.

Patton, S. (2017, September 28). *The Definitive Guide to Playtest Questions*. Schell Games. https://www.schellgames.com/blog/the-definitive-guide-to-playtest-questions

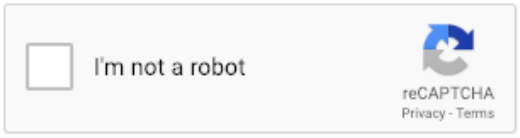
A.5 Posttest & PXI Questionnaire

Email Verification

Thank you for participating in the playtest session! Your feedback is greatly appreciated. Now, if you have a few minutes to fill out the posttest. This test is two sections, the first being about your experience playing the game (11 questions), and the second is about the immune system (13 questions).

But first, please enter the email address where you received the link to this survey to ensure that we know who completed this test.

Please show this robot that
you are not a robot.



The following questions are to capture your experience with the game without the investigator's presence influencing your answers. **So be brutally honest in your answers**. Seriously, no feelings will be hurt. Your truth is requested in how you answer the following statements...

Player Experience

Playing the game was meaningful to me.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I wanted to explore how the game evolved.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I felt I was good at playing this game

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I felt free to play the game in my own way.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I was immersed in the game.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I could easily assess how I was performing in the game.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I liked the look and feel of the game.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

The game was not too easy and not too hard to play.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

It was easy to know how to perform actions in the game.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

The goals of the game were clear to me.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

I had a good time playing this game.

Strongly Disagree	Disagree	Somewhat Disagree	Neutral	Somewhat Agree	Agree	Strongly Agree
○	○	○	○	○	○	○

Transition

Thank you for your honesty. Now for the last section, the immune system.

As before, please do not search for the answers online or elsewhere.

Immunity Quiz

Select the correct statement about innate and adaptive immunity.

- ○ Innate immunity involves cells bearing receptors that are highly specific for a pathogen.
- ○ The first response to a pathogen involves immunological memory.
- ○ Hematopoiesis occurs in the lymph node.
- ○ Phagocytosis involves the uptake and killing of microbes.
- ○ Once lymphocytes mature, they remain in lymphoid organs and do not reenter the blood.
- ○ Don't Know

The first line of defense against microorganisms that infect the body is referred to as _____ immunity.

- opportunistic
- innate
- adaptive
- primary
- central
- Don't Know

When macrophages secrete _____, an inflammatory response ensues.

- lysozyme
- defensins
- lymph
- sebum
- cytokines
- Don't Know

The thin layer of cells that makes up the interior lining of the blood vessels is called the _____.

- mucosa
- epithelium
- endothelium
- connective tissue
- lymphoid tissue
- Don't Know

Which of the following complement components is an opsonin that binds to complement receptor 1 (CR1) on macrophages.

- C3b
- C3a
- Bb
- Ba
- C3bBb
- Don't Know

The name given to cytokines that recruit cells to move toward areas of inflammation is _____.

- chemokines
- caspase-recruitment domains (CARDs)
- inflammakines
- adhesion molecules
- pyrogens
- Don’t Know

What is the name given to the intracellular vesicle in macrophages that have a concentration of degradative enzymes to completely degrade the pathogen?

- Opsonome
- Membrane-attack complex
- Lysosome
- Phagosome
- Phagolysosome
- Don’t Know

What is the function of the C3A protein in the immune system response? What

does it do? (Leave blank if you cannot answer.)

What is the function of the M2 Macrophage in the immune system response? (Leave blank if you cannot answer.)

What happens during inflammation? (Leave blank if you cannot answer.)

What is a pathogen and why is it a concern to the human body? (Leave blank if you cannot answer.)

Review

How confident are you in your answers?

Which questions did you find most difficult?

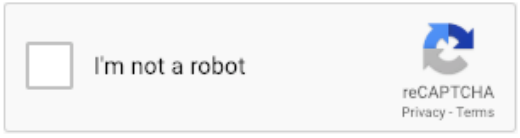
A.6 Delayed Posttest Questionnaire

Email Verification

Welcome to the delayed posttest! It's been 3 weeks since you completed the last posttest, so here is the final posttest. This survey is only the immune system questions, so it should be quick.

Please enter the email address where you received the link to this survey to ensure that we know who completed this test.

Please show this robot that
you are not a robot.



Before we start, can you share whether you played the game since the time you completed the posttest?

- ○ I did not play the game since the posttest.
- ○ I played the game a little since the posttest.
- ○ I played the game a lot since the posttest.

○ [ ] Other

Please share any thoughts you had of the game with the additional playtime.

As before, please do not search for the answers online or elsewhere.

Immunity Quiz

Select the correct statement about innate and adaptive immunity.

- ○ Innate immunity involves cells bearing receptors that are highly specific for a pathogen.
- ○ The first response to a pathogen involves immunological memory.
- ○ Hematopoiesis occurs in the lymph node.
- ○ Phagocytosis involves the uptake and killing of microbes.
- ○ Once lymphocytes mature, they remain in lymphoid organs and do not reenter the blood.
- ○ Don't Know

The first line of defense against microorganisms that infect the body is referred to as _____ immunity.

- opportunistic
- innate
- adaptive
- primary
- central
- Don't Know

When macrophages secrete _____, an inflammatory response ensues.

- lysozyme
- defensins
- lymph
- sebum
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- Don't Know

The thin layer of cells that makes up the interior lining of the blood vessels is called the _____.

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- Phagolysosome
- Don’t Know

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does it do? (Leave blank if you cannot answer.)

What is the function of the M2 Macrophage in the immune system response? (Leave blank if you cannot answer.)

What happens during inflammation? (Leave blank if you cannot answer.)

What is a pathogen and why is it a concern to the human body? (Leave blank if you cannot answer.)

Wrap-up

How confident are you in your answers?

Which questions did you find most difficult?

A.7 Player Performance Rubric

Topic	Description	Poor 0 pts	Fair 1 pt	Good 2 pt	Great 3 pts
Maneuvering	Rate the player's level of moving and evading	Took damage often due to not evading, movement didn't show a clear objective or purpose.	Attempted to avoid pathogens and move with intent.	Avoided pathogens and was fully in control.	Avoided pathogens even in close calls, moved with grace and perfect precision.
Aiming	Rate the player's level of aiming	Missed most shots.	Hit about half of the shots.	Most shots hit. Some long range shots or more difficult shots.	Perfect or near perfect accuracy. Fancy shots from afar and betwixt obstacles
Collecting and Firing C3	Rate the player's ability to collect and fire C3.	Repeatedly failed to collect and fire when needed.	Collected and fired some but often was caught being empty when they needed it.	Collected and fired effectively (wasn't ever empty when needed), but maybe focused too much time on collecting than doing.	Perfectly balanced collecting, firing, and doing other tasks.
Canceling Infection	Rate the player's ability to strategically aim at a pathogen that is actively infecting and canceling it with a C3 hit.	Didn't try.	Attempted.	Made at least 1 cancel shot.	Made every cancel shot.
Collecting C3A	Rate the player's ability to collect C3A.	Repeatedly failed to collect when needed.	Collected some but not enough or effeciently.	Collected effectively but gave too much time or focus on the task.	Perfectly balanced collecting and doing other things.
Macrophage Grab	Rate the player's ability to grab onto pathogens	Never tried.	Tried but failed.	Successful grabs with some misses	Never missed
Phagocytosis	Rate the player's ability to phagocytosis	Never tried.	Tried but failed.	Failed at least once but was successful at least once	Succeeded every time.
M2 Healing	Rate the player's ability to M2 heal	Never tried.	Missed most of inflammation and cells	Healed most inflammation and cells	Full restoration
Winning	Rate the player's ability to win the level	Never won and wasn't ever close.	Almost won.	Won with some challenge.	Won while making the level look way too easy.

A.8 Retrospective Grading Tables

Table 7: Retrospective Grading: P1-P4

Player	Retrospective	Endo	C3	Patho	C3B	C3A	Macro	Phago	Infla	Cyto	Chemo	M2	Total
P1	I think the loop [is] attaching these C3 proteins to Pathogens. And then calling in the Macrophage to come and eat. You can see how this could get out of hand if the immune system doesn't work fast enough, being that it's like, you know, replicating the way that viruses do. A Macrophage eats these things. It does that based on C3 protein. The C3A that was leftover, right? That kind of marks it, which then allows the Macrophage to grab onto it. And then the C3B, that comes off as the thing that attracts the Macrophage.	0	1	1	0.5	0.5	1	0.5	0	0	0	0	4.5
P2	I'm controlling this thing. I think these are red blood cells. And a pathogen influenza virus attacked. Well, the first one was from a vaccine, so it wasn't moving. And collected CB3 proteins to attack the virus. And then I gained C3A, the pink things. And then once I collected six of them, I attracted a macrophage, which would then allow me to phagocytize; to eat the virus. And then two more attacked. Oh, wait. And then I used the M2 to heal the cells. And then two more.	0	0.5	1	0	1	0	1	0	0	0	1	4.5
P3	I think it's the NanoDrone attacked the macrophage. And you collect the yellow bullets of C3A. And, and the last, it ...actually, I don't remember the name of the purple thing. The goal is to absorb the macrophage. And clear that out.	0	0	0	0	0	0	0	0	0	0	0	0
P4	Overall, I'm like a little nano bot that can detect viruses by itself. And then it attaches C3 protein to attract more immune cells and macrophages can detect that C3 protein. So, once they are tagged I can also control the macrophage when they arrive. And then I can kind of like phagocytose the virus by attaching to its C3 proteins. And then after that the cytokines are released and then that increases inflammation. And then like blood elements kind of flow into the space and then M2 macrophages come, and it heals and closes the inflammation. I think that is overall what happened.	0	1	1	0.5	0.5	1	1	1	1	0	1	8

Table 8: Retrospective Grading: P5-P6

Player	Retrospective	Endo	C3	Patho	C3B	C3A	Macro	Phago	Infla	Cyto	Chemo	M2	Total
P5	You're on a sheet of cells and you're this guy, and you pick up little immunity C3s and you become–I kept calling everything by not it's real name. By it's like purple thing or, cause I don't remember any of the names. I think it was psyto-something was the purple thing. And you defeat the pathogen that way. Uh, either by when you fire the little immunity things out then you can latch on to it. And then, oh, at the end I got to be a magic healer of the inflammation and the cells.	0.5	0.5	1	0.5	0	0.5	0.25	0	0	0	0.5	3.75
P6	So we had endothelial cells on which viruses landed from... Actually it's interesting cause the virus is influenza. No, it shouldn't land on the skin, so it should be internal. So I think you got it's breathes inside and it's somewhere inside the lungs, I think, at this point. And then it's the endothelial cells. It's like and then we are acting as this cannon that can shoot C3B receptors to mark and also kind of release this pathogen killing opsinin thing. And then as we shoot that each time, it creates another complement protein that which we collect and then alert the macrophage to enter the scene. And then that macrophage eats the virus and then releasescytokines and antigens into the bloodstream mechanism, which are used by the immune system to like learn or like remember future pathogens. Then we played a tutorial where we interacted with Alice, who explained these steps. Oh yeah, then there was inflammation in the region, and then we get an M2 macrophage. That comes to like heal those cells. And like pull down the inflammation. So, yeah. Oh yeah. And then what happened next, in the game, like we played different levels where we have like multiple viruses and stuff.	1	0.5	1	0.5	0.5	1	0.5	1	1	0	1	8

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