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Visible Light and Its Influence on the Embryonic Viability of the Cricket *Acheta domesticus*

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Abstract

During *in vitro* fertilization, human embryos are incubated without light, and these conditions do not ensure embryo survival. This study explored whether environmental conditions can influence the embryo viability rates of the house cricket, *Acheta domesticus*. In particular, the experiment tested what colors of visible light provide the best incubation conditions to ensure cricket embryo viability. The concept was to use house cricket embryos to represent human embryos. Cricket embryos were chosen as their eggs have soft outer membrane casings and resemble human embryos during the first few days after fertilization. During the experiment, the adult crickets laid their eggs into one of six soil-filled boxes called substrates. Each substrate was placed into one of six storage containers filled with adult crickets and lit with a different colored visible light (red, yellow, green, blue, white, or no light). After two days of breeding, the egg-filled substrates were removed from the adult crickets and placed in another storage container of the same color light. After incubation under heat-emitting lamps and under one of six light colors, nymphs were counted after hatching to determine embryo viability. After three trials, the red light provided the significantly highest viability rate, with yellow and no light being comparable seconds. The green, blue, and white lights showed significantly lower viability rates than no visible light. My results raise the speculation that exposing fertilized mammal eggs to visible light colors might have the same effects during the *in vitro* fertilization process.

Keywords

Embryology, Embryo, Embryo Viability, Embryonic Viability, Viability, Viability Rate, Incubation, *In Vitro* fertilization, *In Vitro*, Fertilization, Visible Light, Cricket, Acheta *domesticus*

1. Introduction

Does visible light influence the embryonic viability of the cricket, *Acheta domesticus*? Research has proven that variables such as temperature, space, food, and humidity have an effect on all living organisms. The *Acheta domesticus*, more commonly known as the house cricket, is no exception. The color of visible light is one variable that has an effect on living organisms including the cricket. The goal of this research project was to determine whether visible light had an influence on cricket embryo viability. The hypothesis of this project states the following: If embryos are placed under visible light, the visible light will have an influence on embryonic viability. The color of visible light was the independent variable addressed in this project. During the process of this research, it was determined which visible light color had a positive influence and which had a negative influence on cricket embryo viability. To reach this goal, adult crickets were placed in man-made habitats for breeding and were provided with a designated location to lay their eggs. The eggs were removed from the adult crickets, incubated under one of six visible light colors, and monitored until they hatched.

Human, insect, and all other animal embryos are similar to one another during the early stages of embryonic development [1] [2]. According to research, the cricket *Gryllus bimaculatus* has the most advanced genetic makeup than any insect [3]. During the beginning stages of embryo development, the embryonic cells transition through a cleavage stage and then pull together toward the periphery of a blastoderm to form the embryo and protective membranes of the cricket [3]. The same is true for human embryonic development. The embryonic cells transition through a cleavage stage of cell mitosis and then travel to form the periphery of the blastocyst [4] [5]. The extraembryonic cells that line the cricket blastoderm form two layers called the amnion and serosa layers [2] [3]. The inner layer called the amnion, forms the tissues that provide containment of the fluid within the amniotic cavity [2]. The outer layer, the serosa, gives rise to the protective barrier of the entire egg [2]. The blastocysts of human embryonic development are composed of two layers as well. The inner cellular layer becomes the fetus and the outer layer, the trophectoderm, becomes the placenta [5]. If visible light influences embryo viability in crickets, the same may be true with human embryo viability when visible light conditions are controlled during the *in vitro* fertilization (IVF) process.

Crickets were chosen as the experimental unit for this research experiment as cricket embryos hold similar properties to human embryos [2]. "Crickets are hemimetabolous, (and) unlike the indirectly developing holometabolous insects, they do not pass through a larval stage nor go through total metamorphosis" (p. 141) [3]. This facilitated the counting process of the viable embryos. Nymph crickets also come from soft egg shells that allow for light penetration [2] [3]. The same holds true with the zona pellucida, the transparent covering of the human blastocyst when exposed to light during the IVF process [6]. In addition, adult female crickets are capable of laying eggs daily as they store deposited stores

of sperm in their bodies for up to 10 - 12 days and until substrates become available to lay their eggs. Since adult female crickets lay eggs daily, the eggs can be collected two days after being introduced to a soil substrate. The incubation period for cricket embryos is 11 - 14 days allowing the research project to be completed within a reasonable time frame. Fertilized cricket eggs are naturally deposited into the darkness of soil as fertilized human eggs are naturally fertilized in the darkness of the uterus. This research project exposes cricket eggs to light immediately after fertilization in the same way that human eggs are exposed to light during IVF.

IVF has been around for decades. "Between 1878 and 1953, numerous attempts were made to fertilize mammalian eggs *in vitro*. Fertilization is a progressive, continuous process, beginning with the penetration of an egg by a spermatozoon, which results in the extrusion of the second polar body from the egg" (p. 182) [7]. Throughout the IVF process, doctors implant multiple embryos into the uterus after a few days of *in vitro* incubation in hopes that one egg will survive. "(IVF) Accounts for millions of births worldwide and 1% - 3% of all births every year in the U.S. and Europe" (p. 156) [8]. In 2004, a study showed that 3.6% of women younger than 35 years of age had a live birth after being implanted with an average of 2.5 embryos. As a result, 32.7% of these women delivered twins and 4.9% delivered triplets [8]. Therefore, improvements in the IVF process are needed in order to reduce multiple births that pose risks to both the mother and baby. With the further development of IVF, IVF has become increasingly popular despite its risks. Demands for new technology to better the process of IVF have resulted.

There are similar studies to this research, but those studies have slightly different procedures and outcomes. In one study, these researchers studied IVF and noted, "Damaging effects of visible light primarily concentrate to the 400 - 500 nm range. Therefore, the stress upon the embryos can be reduced by mounting filters on the inspection microscopes such that radiation energy in the 400 - 500 nm range is excluded" (p. 102) [9]. This group of researchers found that blue visible light has the potential to damage embryo cells. They failed to discuss, however, the viability of the embryos. Another study concluded that exposure to ambient visible light during IVF damages the cells of embryos by changing biophysical impedance (BI) [10]. Likewise, this group of researchers focused on the effects of cell growth using blue, green, red, and white lights, and not on the viability of the fertilized egg. Last, a third study concluded that hatchability, embryo weight, brain weight, and BI were altered by exposure to the light colors of red, blue, and green lights [11]. Chicken eggs were used in this study rather than cricket eggs. Chicken eggs have hard and opaque shells that filter out visible light to protect the chicken embryos when exposed to light in their natural environments [11]. Human and cricket embryos have soft membrane casings and they are naturally incubated in the darkness. The color of visible light and its influence on embryo viability is a topic with very little research. This research project addressed the possible effects of cricket embryo viability when exposed to the

colors of visible light during the incubation period of embryo development. The findings of this study could lead researchers to delve deeper into similar research studies to determine what color of visible light would allow for an increased rate of human embryo viability during the IVF process.

2. Materials and Methods

2.1. Introduction

The true reason for studying cricket egg viability is to determine whether the results of the study can be beneficial for further research on the human IVF process. According to research, the human embryo survival rate is higher when the embryo is implanted on the fifth day after fertilization as compared to the third day [5]. Some reproductive endocrinologists will implant the embryo into the uterus on the third day after egg fertilization knowing that the egg may not survive in the *in vivo* conditions. Often, the embryo fails to adhere to the uterine wall as the embryo size is too small. Reputable reproductive endocrinologists implant embryos after the fifth day of fertilization when the embryos are going through the blastocyst stage [5]. During this time, the embryo becomes larger and adheres to the uterine wall better than when it is implanted on the third day. Storing the embryo for five days in the laboratory requires reproductive endocrinologists to set up the best conditions for the survival of the embryo. According to Hong, "About 80 percent of eggs will fertilize (day 1 success rate), and of those about 30 - 50 percent will make it to the blastocyst stage (day 5 or 6)" [5]. This equates to an overall survival rate of 24% to 40%. Bettering the storage conditions with the right visible light may improve the odds of embryo viability to the fifth day. With the results of this experimental study, my goal was to determine whether or not visible light influences embryo viability. In order to achieve valid results, numerical data collection was required to show the difference in embryo viability for each color condition of visible light. This involved counting the amount of viable nymph crickets in each colored box.

2.2. Methodology and Process

This research project is of a quasi-experimental observational control group time-series design that uses QUAN + qual data. In order to show significant differences in the number of crickets that hatched under various light conditions, quantitative data on the number of viable crickets was obtained. This data was used in a one-way ANOVA to calculate the probability that the null hypothesis was not true and to validate the research. Furthermore, qualitative data was used to support the quantitative data. The control-group, time-series design was used as multiple groups were studied over a period of time which included blue, green, white, yellow, red, and no visible light conditions. These colors were chosen as they are the most basic colors. The no visible light group represented the conditions that are set for human embryo cryostorage. The no visible light testing condition was the one group that did not receive the treatment of any visible

light. The white visible light also represented the ambient light that embryos are exposed to through the *in vitro* fertilization process. The control variables were kept consistent making any confounding variables low and less likely to skew the outcome of the dependent variable.

2.3. Environment Setup

The set-up of the materials and equipment is shown in Figure 1 and Figure 2. For this study 12 storage containers measuring 24"L × 16"W × 15"H in dimension were used to house the crickets. A 14"L × 9"W square was cut out of the

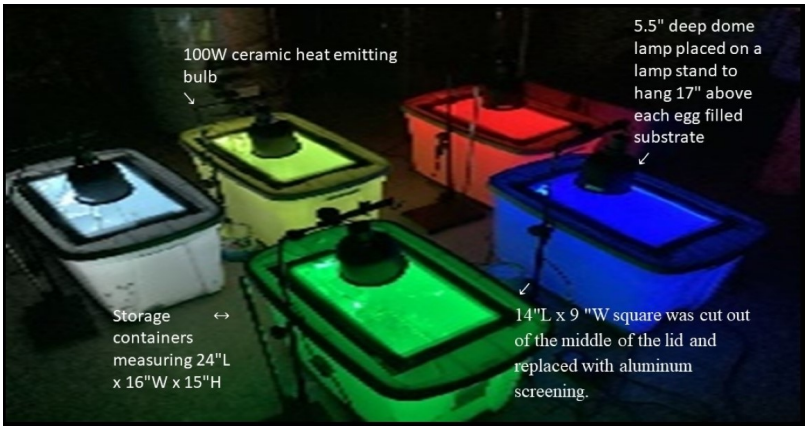

Figure 1. Exterior view of the testing environment. Note: The six containers were used as incubators for cricket embryos. Before the eggs were incubated the crickets were given two days to lay their eggs. Silhouettes of crickets can be seen through the containers. The container without light is in the background. Picture taken by Matthew Ferenz.

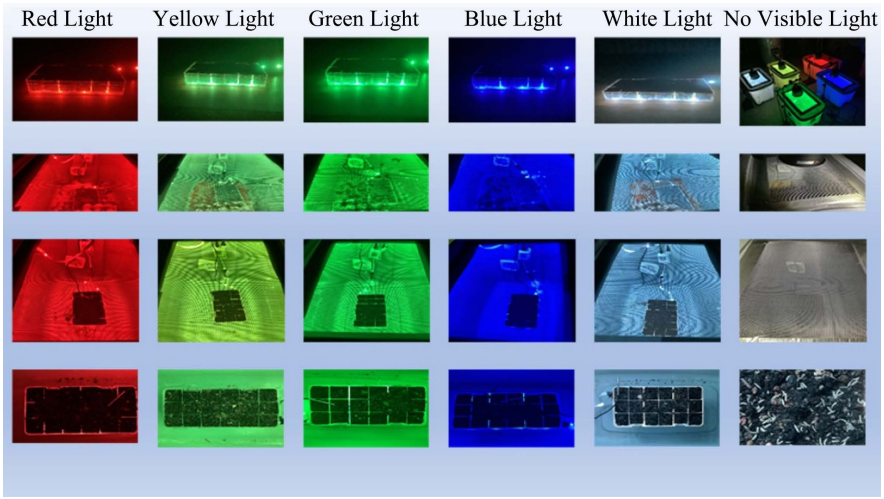

Figure 2. Interior view of the testing environment. Note: The photographs in these pictures show how the equipment noted in Figure 1 was functionally used during the experiment. The first column shows the unused LED substrates. The last cell of the first column displays the cricket-filled storage containers. The second column shows the adult cricket breeding environment and the third column displays the embryo incubation environment. Egg-filled substrates are depicted in the last column. Pictures taken by Matthew Ferenz.

middle of the lid and replaced with an aluminum screening. Duct tape and packaging tape held the screening into place from the outside of the lid. Sixty-watt light-emitting diode (LED) strips (17.5 feet of 90 diodes) were adhered to the inside parameter of the lid surrounding the screening. Six containers bedded with vermiculite were assigned a color light condition of either red, yellow, green, blue, white, or no light. Each container was supplied with 500 randomly picked adult crickets until they laid their eggs into one designated substrate. The six substrates were housed in clear tackle boxes measuring 6.77"L × 3.94"W × 0.87"H with an inside grid size dimension of 6.57"L × 1.18"W × 0.79"H. The substrates were filled with topsoil and covered with aluminum screening to prevent the adult crickets from eating the eggs. Sixty-watt light-emitting diode (LED) strips (17.5 feet of 90 diodes) were adhered to the base of the substrate containers. The light emitted from the strips provided color to the soil as the light was transmitted and emitted from the plastic dividers of the clear plastic tackle box. The visible light color of the substrate was chosen to match the storage container that houses it. After two days of laying eggs, the substrates were transferred to a second storage container with the same color of visible light where the cricket eggs would eventually hatch. A 100 W ceramic heat emitting bulb will be used in a 5.5" deep dome lamp and placed on a lampstand to hang 17" feet. This bulb emitted no visible light. Each container contained a digital thermostat and temperature controller to regulate the heat between 83 to 89˚F. The probes of the thermostats were placed on each of the substrates. A hygrometer to measure humidity was placed in each box as well. Orange slices and water from a spray bottle were used in controlled amounts. Adult crickets were ordered by mail and delivered in six boxes of 500 crickets each and were ready to breed. Each box of crickets was placed into one 18-gallon storage container.

2.4. Data Sources and Sample

The experimental units for this experiment were chosen for a non-random grouping sampling design. The crickets were chosen non-randomly. Adult crickets were chosen as they were at the age where they could lay eggs. Crickets were also chosen because they don't go through a larvae stage and have a soft shell. The crickets were found online and could be bought according to the age and number of crickets needed for the experiment. Therefore, all of the adult crickets in each box were the same age, genus, and species. Five hundred crickets were used to eliminate any sampling errors. The large number of crickets in this experiment was used to ensure that the ratio of male-to-female crickets in each box was close to the ratio of male-to-female crickets that occur in nature.

Although it is difficult to have a bias in this research experiment, variables were controlled to ensure equal conditions for each testing environment. This included keeping the same ranges of temperature, ranges of humidity, numbers of lights, and amounts of food. In order to keep the procedures consistent, data was collected twice each day. Bias in my research is less likely as it doesn't pertain to hu-

mans and is strictly to find viability under different light conditions. Three trials were implemented to ensure the reliability of the collected.

2.5. Instruments Used for Data Collection

In order to obtain the results, specific instruments were used for measuring and recording data. In order to obtain the information, data was collected on Google Sheets computer software spreadsheets. Temperature and humidity measurements were conducted and recorded twice a day for each box. Humidity was monitored through analog hydrometers placed inside the testing containers. Temperatures were regulated with digital thermostats which were connected to the heat lamps to ensure that the temperatures for each condition remained between 83˚ - 89˚F. One thermometer probe was placed on the substrates for each box to ensure that temperature measurements were taken from the same distance from the substrates to the heating lamps. Equal amounts of water were sprayed onto the substrates to keep the humidity levels equal and the soil moist. Ceramic heat emitters (100 watts) were used for temperature control and placed the same distance from the substrates for each testing environment. Sixty-watt light-emitting diode (LED) strips (17.5 feet of 90 diodes) were adhered to the inside parameter of the lid surrounding the screening. Sixty-watt light-emitting diode (LED) strips (17.5 feet of 90 diodes) were also adhered to the base of the substrate containers. The light emitted from the strips provided color to the soil as the light was transmitted and emitted from the plastic dividers of the clear plastic tackle box. These LEDs did not emit heat. Nymph crickets were hand-counted after they hatched to determine embryo viability.

2.6. Data Analysis

Upon completion of each trial, a comparison of the number of nymph crickets for each visible light condition was conducted through statistical analysis. The outcome of the quantitative data and qualitative data has been provided in the Results section.

3. Results

3.1. Cricket Viability

The number of viable cricket embryos for the three trials of each color light is shown in **Figure 3** and accompanied by the standard deviations of viable cricket embryos for the projected populations. As shown in **Figure 3**, the largest number of viable cricket embryos among each visible light group was found in the red visible light group (Trial #1 = 51, Trial #2 = 46, and Trial #3 = 72). As indicated by the use of error bars, the number of crickets counted in Trial #1 and Trial #2 of the red visible light sample groups, fell within the red visible light standard deviation of ±13.08, while Trial #3 fell slightly above one standard deviation. The white visible light treatment rendered no viable cricket embryos, therefore having a standard deviation of zero. The blue visible light group was

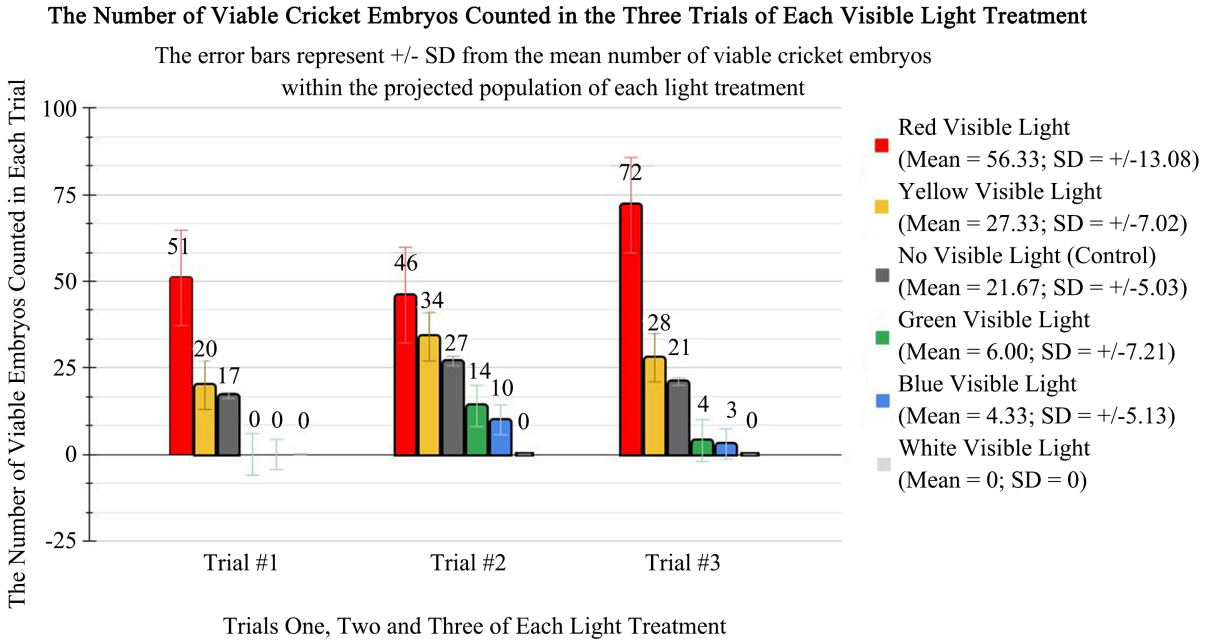

Figure 3. Number of viable cricket embryos counted in the three trials of each visible light treatment. Graph created by Matthew Ferenz.

found to have the second lowest number of viable cricket embryos among each visible light group (Trial #1 = 0, Trial #2 = 10, and Trial #3 = 3). The number of crickets counted in Trials #1 and #3 of the blue light color fell within the standard deviation of the mean (±5.13), while Trial #2 fell slightly above one standard deviation of the mean. The green visible light group was similar to the blue visible light. The number of crickets counted in Trials #1 and #3 fell within the standard deviation of the mean (±7.21), while in Trial #2 each of the green and blue colors fell slightly above one standard deviation of the mean. The yellow visible light group and the no visible light group were comparable to each other in that Trials #1, #2, and #3 fell within the standard deviation of the mean (yellow light $\sigma = \pm 7.02$; no Light $\sigma = \pm 5.03$).

In addition to comparing the standard deviation with the number of viable crickets for the three trials of each light color, the mean number of crickets calculated from the three trials for each light color was also compared to the mean of the mean number of crickets calculated from the three trials for each visible light color. **Figure 4** shows the standard error from the mean through the use of error bars. As shown in **Figure 4**, the mean number of viable crickets for the red visible light color was an outlier from the rest of the visible light colors with a mean of 56.33 and a standard error from the mean of ±7.97. The yellow and no visible light groups were comparable to each other with means of 27.33 and 21.67 and with standard errors from the mean of ±4.05 and ±2.90 respectively. The green and blue visible groups were lower in the mean number of viable crickets (green light $\bar{x}$ = 6.00; blue light = 4.33). The standard error of the mean for the green visible light was ±4.16 and overlapped the blue visible light standard error of the mean of ±2.96.

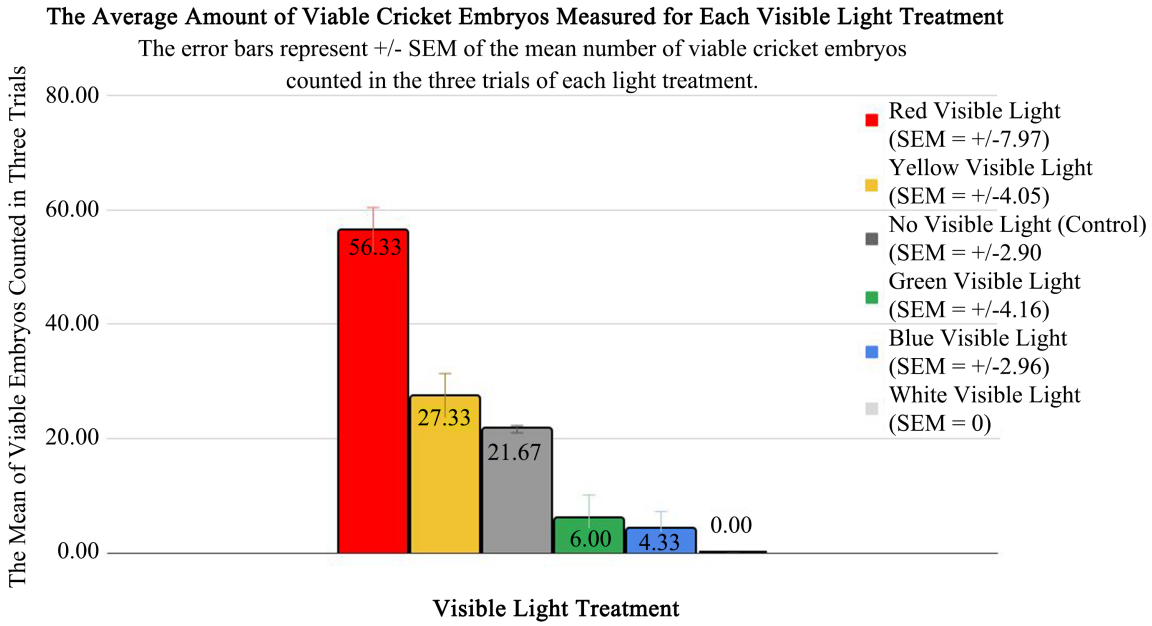

Figure 4. The average amount of viable cricket embryos measured for each visible light treatment. Graph created by Matthew Ferenz.

A one-way ANOVA test was used to test the hypothesis that visible light influences the embryonic viability of the cricket *Acheta domesticus*. The one-way ANOVA was used to compare the influence of visible light (recorded as red, yellow, green, blue, white, or no light) on cricket viability. The one-way ANOVA revealed a statistically significant difference between the various colors of visible light and the viability of cricket embryos ($F(5, 12) = [23.23]$, $p = 0.0000087$). This number should be under 0.05 and the results do so which is validating my research.

3.2. Temperature and Humidity

This study was conducted in three trials. With each trial temperature (˚F) and humidity measurements were recorded twice a day. The average calculations and measurements of variability were obtained from the data. As shown in **Figure 5**, the highest temperature for the mean of all three trials was found with the white visible light with a temperature of 85.81˚F, and the lowest temperature for the mean of all three trials was found with no visible light at 82.52˚F.

The standard error of the mean for the mean temperatures taken in all three trials for each visible light was also calculated and represented in **Figure 5** with error bars. The standard error of the mean for the mean temperatures taken in all three trials for the yellow visible light was ±1.34, for the white visible light was ±1.38, for the red visible light was ±1.40, for no visible light was ±1.45, for the green visible light was ±1.48, and or the blue visible light was +1.56. The standard error bars of **Figure 5** show no significant differences in temperature measurements between the visible light colors, statistically indicating that the differences in temperature measurements had no effect on embryo viability.

Coupled with temperature measurements, humidity measurements were also recorded twice a day to keep the controls consistent. The average calculations and measurements of variability were obtained from the data. As shown in Figure 6, the highest humidity measurement for the mean of all three trials was found with the no visible light treatment with a humidity measurement of 27.11%, and the lowest humidity measurement of the mean of all three trials was found with the blue visible light with a humidity measurement of 23.47%.

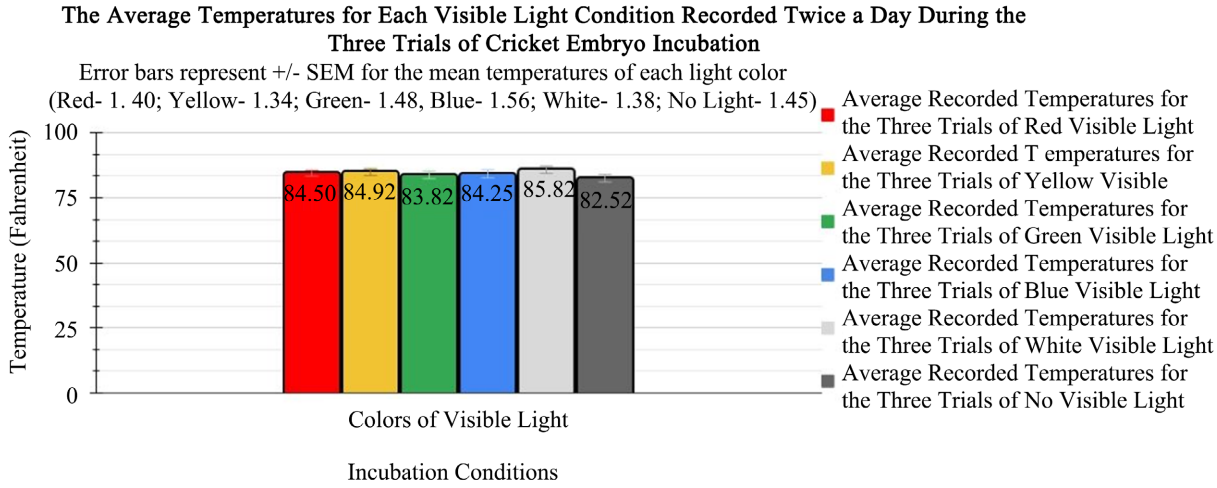

Figure 5. The average temperature for each visible light condition recorded twice a day during the three trials of cricket embryo incubation. Note: The standard error bars show no significant differences in temperature measurements between the visible light colors, statistically indicating that the differences in temperature measurements had no effect on embryo viability. Graph created by Matthew Ferenz.

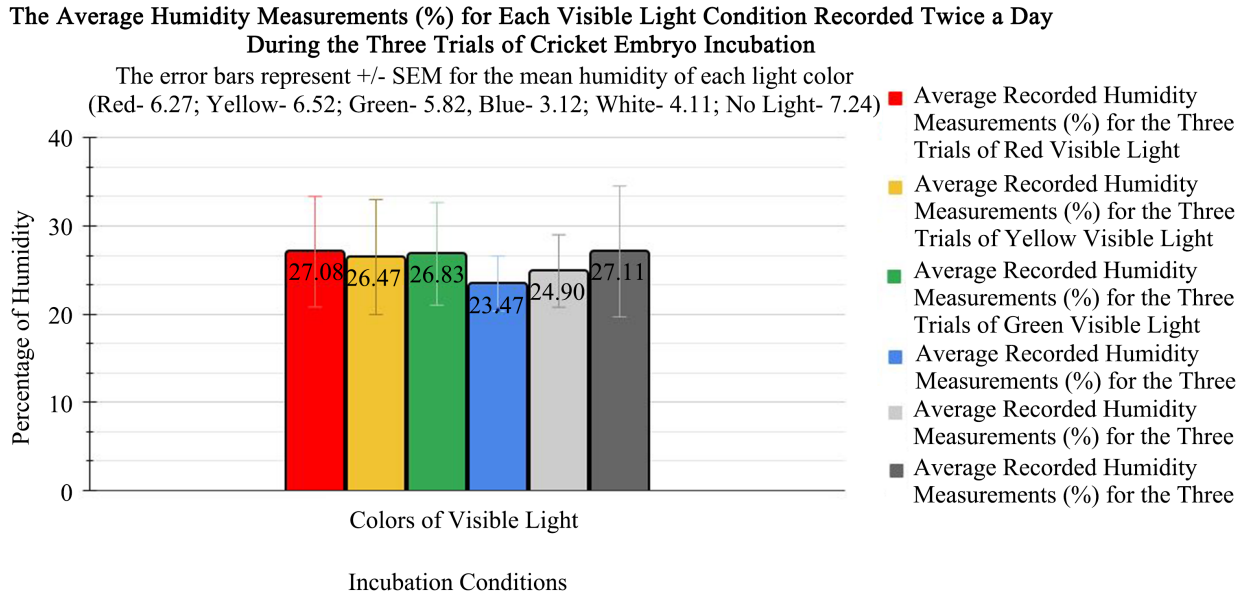

Figure 6. The average humidity measurements (%) for each visible light condition recorded twice a day during the three trials of cricket embryo incubation. Note: The standard error bars show no significant differences in humidity measurements between the visible light colors, statistically indicating that the differences in humidity measurements had no effect on embryo viability. Graph created by Matthew Ferenz.

The standard error of the mean for the mean humidity measurements taken in all three trials for each visible light was also calculated and represented in **Figure 6** with error bars. The standard error of the mean for the mean humidity measurements taken in all three trials for the blue visible light was ±3.12, for the white visible light was ±4.11, for the green visible light was ±5.82, for red visible light was ±6.27, for the yellow visible light was ±6.52, and for no visible light treatment was ±7.24. The standard error bars of **Figure 6** show no significant differences in humidity measurements between the visible light colors, statistically indicating that the differences in humidity measurements had no effect on embryo viability.

4. Discussion

4.1. Highest Embryonic Viability

After conducting three trials on the six visible light test groups, red visible light had a higher embryo viability rate than any other group as determined by the most nymphs counted. With this data, it is safe to say that the red visible light conditions may have a positive influence on cell growth. In fact, there are medical uses for red visible light known as red light therapy. Red light therapy is used on humans to improve skin appearance for conditions such as redness, acne roughness, and/or wrinkles [12]. A group of people were tested with red light therapy, and the research found that the treated subjects experienced significantly improved skin complexion, skin sensation, skin smoothness, and collagen density [12]. Within-group comparisons, t0 - t30 of red-light therapy (RLT) and energizing light therapy (ELT) groups show that skin complexion, skin sensation, collagen intensity scores, skin smoothness, and wrinkle status improved significantly with RLT [12]. This evidence supports that red light therapy helps the rebuilding of skin cells. A study found, "The use of LED light sources with 590, 633, and 830 nm wavelengths for athermal light-only photorejuvenation has grown rapidly in recent years. Additional wavelengths have been shown to be efficient in altering cellular functions, such as 570, 620, 680, 760, and 820 nm" (p. 98) [12]. Most of the wavelengths of these readings are of the color red (600 - 700 nm). Red visible light alters cellular function to allow for cellular growth. Red light in my experiment may influence cell growth which increases the viability of cricket embryos. Therefore, results may indicate that there is a correlation between visible red light, cell growth, and embryo viability.

4.2. Lowest Embryonic Viability

After conducting this research on embryo viability results indicate that the embryo rates of the white and blue visible light conditions were the lowest of any of the groups as determined by the least number of nymphs counted. Of the six color visible lights tested in this experimental research, white had the lowest viability. Many egg embryos were in the soil substrate in the white boxes, but no nymphs hatched from these eggs. As a result, white visible light may have inhi-

bited the nymphs from hatching. A doctor at a fertility clinic stated, "Keep the exposure time as short as possible, e.g. by narrowing the illuminated area of the microscope table, such that only the embryo being inspected is present there" (p. 102) [9]. During IVF, eggs are fertilized outside of the human body and are exposed to ambient light when they are being monitored for growth and viability in the lab. During typical IVF lab procedures, embryos receive radiation energy from light. After exposure to radiation energy for a certain time range, the radiation begins to stress the biological systems [9]. As a result, white light would not be a recommended visible light color to use when storing embryos during the process of IVF. Because white light damages cells in IVF, it may explain why no nymphs hatched under the white visible light conditions.

4.3. Lowest Non-White Embryonic Viability

White visible light was determined to have the lowest rate of cricket embryo viability. White visible light is composed of all colors of the visible light spectrum and will have influences on cell growth. Blue light is in the 400 - 500 nm range of the light spectrum. The medical field uses blue visible light in the form of photodynamic therapy (PDT). Blue fluorescent light is applied to the skin to kill cancer cells. In addition, blue light is also used to kill bacteria from phones and toothbrushes using blue light cleaners. Blue light is also used for teeth whiteners at the dentist's office. Past research states, "A typical *in vitro* fertilization procedure implies 400 - 500 nm irradiance doses and such irradiation doses contribute to the number of non-lethal stress factors related to *in vitro* culture, according to the data, possibly through stress on the respiratory chain. Reducing stress on the mitochondria during *in vitro* culture, in order to increase oxidative capacity, could be helped by a general reduction of light exposure" (p. 101) [9]. Not only does blue visible light destroy cell growth during IVF, but blue visible light can inhibit growth in other areas. "It has been found that excessive blue light exposure, especially at night when melatonin production peaks, can not only damage the retina through the ocular surface. It can also stimulate the brain, inhibit melatonin secretion, and increase corticosteroid production, thereby destroying hormonal secretion and directly affecting sleep quality" (p. 2001) [13]. As a result of these studies, it is safe to say that blue visible light breaks down cell growth. This may be the reason why the blue visible light yielded low viability rates of cricket embryos.

4.4. The Visible Light Spectrum

Exposure to visible light color influences cricket embryo viability rates that are illustrated in **Figure 7**. In **Figure 7**, red light is the furthest to the left on the visible light spectrum and has the highest viability rate. Blue is the furthest to the right on the visible light spectrum and has the lowest viability rate of all of the color visible lights. Chances of embryo viability become less as the visible light colors move to the right on the visible light spectrum. The control group, which had no visible light and is used in IVF storage ranked between the yellow and

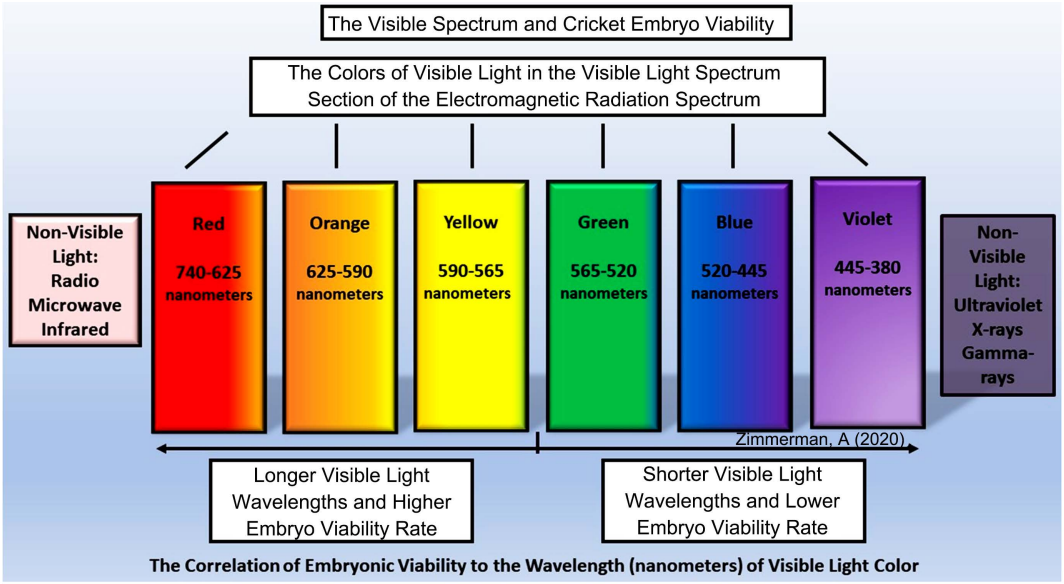

Figure 7. The visible spectrum and cricket embryo viability [14]. Graph created by Matthew Ferenz.

green for the embryo viability. The red and yellow visible lights had greater embryo viability rates compared to what is being used now with dark storage. Therefore, during the IVF process, the human embryo viability rate may increase when embryos are stored in red or yellow visible light. The red and yellow visible lights also had greater embryo viability rates compared to the white visible light. When exposing the human embryo to laboratory white light, it may be beneficial to use rooms lit with red or yellow lighting or to use instruments or equipment that omit yellow or red light rather than white light. Blue light conditions also resulted in a low viability rate in this research experiment. Blue light should also be avoided in IVF laboratories. Embryos should not be exposed to the blue light of computers, cell phones, or medical equipment. This blue visible light could compromise the viability of the embryo.

4.5. Limitations

The methods used to obtain the results of this research experiment, however, had minimal limitations. Every color on the visible light spectrum was not tested as there would not have been enough room in the testing area to do so, and the cost of purchasing more materials and equipment would have been too expensive. Another limitation of this research project was the sole subject testing of the house cricket or *Acheta domesticus.* No other insects were tested. Would the same results occur with a different species of crickets or genus of insects? Furthermore, another limitation was in the seasonal timing of cricket breeding. Crickets tend to lay more eggs in the summer months compared to the winter months. The summer is humid and keeps the eggs moist. Winter is drier and doesn't allow for high humidity levels. Despite these limitations, the limitations had little to no effect on the results as the results were significant. With their close similarities to humans, predictions can be drawn for future studies.

4.6. Implications

The results found in this study could be beneficial to reproductive endocrinologists and to other physicians/scientists who carry out IVF. The results show clear differences in the embryo viability rates due to exposure to various visible light conditions. Most laboratories that conduct IVF store the embryos in dark incubators. My research and its findings can question whether storing these embryos in dark incubators is the best option knowing that incubation under red and yellow visible light increases the embryo viability of the cricket.

5. Conclusions

The goal of this research was to address the gap existing in previous research that is egg embryo viability under various colors of light. To address the following gap in past research, the following research question was developed: Does visible light have an influence on the embryonic viability of the cricket *Acheta domesticus*. The following hypothesis was also developed: If embryos are incubated under visible light, the visible light has an influence on embryonic viability.

From the review of research that was completed on this topic of *in vitro* fertilization, predictions were made from the hypothesis. These predictions were similar to the final results of this research experiment. The relationship between visible light and embryo viability was not coincidental. Red light is used for therapy to improve skin appearance, hence, improving cell growth. Likewise, the results of this research experiment also indicated that embryo viability that requires cell growth is greater when using red visible light for incubation. The highest average number of nymphs from all color conditions was 56.33 for the red visible light group. On the other hand, blue light is used to break down bacteria. The results of this research experiment also indicated that embryo viability rates were lower for the visible blue light conditions when using blue visible light for incubation. Blue visible light had the lowest average number of nymphs for all color conditions was 4.33. White visible light had no viable crickets in all three trials. Cricket eggs have a yellow tint and may have reflected yellow and red lights and have absorbed an accumulation of harmful visible light energy found to the right of the yellow visible light color on the visible light spectrum. The control group that had no visible light fell between the yellow and green visible light colors for embryo viability rates. The crickets housed in darkness were deprived of the benefits from the visible light colors found to the left of the yellow visible light and were also sheltered from the harmful energy of the visible light colors found to the right of the green visible light on the visible light spectrum. The white visible light group represents the ambient light emitted in laboratories and the no visible light group represents the storage conditions of the embryos. White light rendered no viable cricket embryos and the no visible light condition did not yield the best viability rate for the cricket embryos. The red light and yellow light conditions showed to be greater viability rates than the white

visible light and no visible light conditions that are used in IVF laboratories today.

Future Research

Further research can be completed by embryology researchers and other professionals in this field. These professionals can use the results from this research experiment to study human embryo viability as it is influenced by visible light colors during the IVF process. If professionals in this field find the best visible light conditions for embryo incubation, multiple eggs would not have to be harvested. Fertilized eggs would be more likely to survive *in vitro* to the fifth or sixth day when implantation of the egg into the uterine is more successful than implantation prior to the fifth day after fertilization. With the results of this research experiment and with future research, laboratories can be modified to include incubation under red or yellow light to increase embryo viability. This would allow embryologists to ensure greater embryo viability rates.

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Conflicts of Interest

The author declares no conflicts of interest regarding the publication of this paper.

References

[1] Hill, M.A. (2023) K12 Human and Other Animal Development. Embryology. https://embryology.med.unsw.edu.au/embryology/index.php/K12_Human_and_Other_Animal_Development

[2] Panfilio, K.A. and Chuva de Sousa Lopes, S.M. (2022) The Extended Analogy of Extraembryonic Development in Insects and Amniotes. *Philosophical Transactions of the Royal Society B*: *Biological Sciences*, **377**, Article ID: 20210268. https://doi.org/10.1098/rstb.2021.0268

[3] Donoughe, S. and Extavour, C.G. (2016) Embryonic Development of the Cricket *Gryllus bimaculatus*. *Developmental Biology*, **411**, 140-156.

https://doi.org/10.1016/j.ydbio.2015.04.009

[4] Jansen, R.P.S. (2005) Benefits and Challenges Brought by Improved Results from *in Vitro* Fertilization. *Internal Medicine Journal*, **35**, 108-117. https://doi.org/10.1111/j.1445-5994.2004.00759.x

[5] Hong, K.H. (2022) How Many of My Embryos Will Make It to the Blastocyst Stage. Reproductive Medicine Associates Network. https://rmanetwork.com/blog/blastocyst-how-many-embryos-stage/

[6] Litscher, E.S. and Wassarman, P.M. (2020) Zona Pellucida Proteins, Fibrils, and Matrix. *Annual Review of Biochemistry*, **89**, 695-715. https://doi.org/10.1146/annurev-biochem-011520-105310

[7] Bavister, B.D. (2002) Early History of *in Vitro* Fertilization. *Reproduction*, **124**, 181-196. https://doi.org/10.1530/rep.0.1240181 https://web.archive.org/web/20200305134243id_/https://rep.bioscientifica.com/downloadpdf/journals/rep/124/2/181.pdf

[8] Eskew, A.M. and Jungheim, E.S. (2017) A History of Developments to Improve *in Vitro* Fertilization. *Missouri Medicine*, **114**, 156-159. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6140213/

[9] Ottosen, L.D., Hindkjaer, J. and Ingerslev, J. (2007) Light Exposure of the Ovum and Preimplantation Embryo during ART Procedures. *Journal of Assisted Reproduction and Genetics*, **24**, 99-103. https://doi.org/10.1007/s10815-006-9081-x

[10] Bennet, D., Viswanath, B., Kim, S. and An, J.H. (2017) An Ultra-Sensitive Biophysical Risk Assessment of Light Effect on Skin Cells. *Oncotarget*, **8**, 47861-47875. https://doi.org/10.18632/oncotarget.18136

[11] Abdulateef, S.M., Al-Bayar, M.A., Majid, A.A., Shawkat, S.S., Tatar, A. and Al-Ani, M.Q. (2021) Effect of Exposure to Different Light Colors on Embryonic Development and Neurophysiological Traits in the Chick Embryo. *Veterinary World*, **14**, 1284-1289. https://doi.org/10.14202/vetworld.2021.1284-1289

[12] Wunsch, A. and Matuschka, K. (2014) A Controlled Trial to Determine the Efficacy of Red and Near-Infrared Light Treatment in Patient Satisfaction, Reduction of Fine Lines, Wrinkles, Skin Roughness, and Intradermal Collagen Density Increase. *Photomedicine and Laser Surgery*, **32**, 93-100. https://doi.org/10.1089/pho.2013.3616

[13] Zhao, Z.C., Zhou, Y., Tan, G. and Li, J. (2018) Research Progress about the Effect and Prevention of Blue Light on Eyes. *International Journal of Ophthalmology*, **11**, 1999-2003.

[14] Zimmerman, A. (2020) What Is the Visible Light Spectrum? ThoughtCo. https://www.thoughtco.com/the-visible-light-spectrum-2699036