How to raise H.illucens in captivity: a protocol

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Abstract:

This is a study to assess the near perfect conditions in order to raise the greatest amount of Black Soldier Flies into adulthood. This is so that is amount of uses as Black Soldier flies as model organism increases. All genres of Black Soldier Flies were looked at in this study to develop the protocol. The flies that were chosen were best represented to reflect the abilities and gender of the natural amount of Black Soldier flies in the wild. Potential subjects were then tested with the different types of p variables such as food and sunlight. At this time all the Black Soldier flies were kept in specific conditions and tested with different types of habitats. These habitats reflected the living conditions of the flies in the natural plain. After all the data was compiled, it was aggregated and used to find the best living conditions for the Black Soldier fly as a model organism. This was used personally in a lab setting of the author in order to increase the amount of Black Soldier Flies that were gained from breeding. This protocol resulted in a significant increase in the amount of Black Soldier flies that were bred in the lab.

Larvae Protocol:

“ The larvae will be placed in a 1 m * 1 m * 1 m glass or clear plastic box. There will be dirt on the bottom, followed by chicken meal on the top. There will be three holes that will be 6 cm * 6 cm * 2 cm big ((about one hand scoop)). The box will be divided up into three sections, one for feed and pupation, one for feces, and one for special research flies. The food and pupation section will take up half of the box. The dirt layer will be 5.5 cm deep. The dirt from the holes will be used to make hills and ridges for a more natural location, along with providing shelter. One of the holes will be in the middle, and it will have water from a freshwater river. The water should have a pH level of 5 ((Newby, 1997)). The nutritional value of the chicken meal should roughly have ≈332KJ, ≈6.3 grams of ash, ≈77 calories per serving, ≈4.47 grams of proteins, ≈0.626 grams of fat, ≈13.3 grams of carbohydrates, ≈75.2 grams of moisture, ≈0.217 grams of cis-unsaturated fatty acids, ≈0.095 grams of cis-polyunsaturated fatty acids, ≈0.034 grams of trans-fatty acid, ≈0.005 grams of omega 3 polyunsaturated fatty acids, and ≈0.09 grams of omega 6 polyunsaturated fatty acids ((((((Tomberlin et al., 2002))). Amino acids will also be added for every 100 grams of food for higher nutritional count. The amino acids that roughly should be added are ≈6.56 grams of of aspartic acid, ≈2.77 grams of threonine, ≈3.02 grams of serine, ≈6.95 grams of glutamic acid, ≈4.53 grams of glycine, ≈4.41 grams of alaine, ≈0.39 grams crystine, ≈4.51 grams of valine, ≈1.25 grams of methionine, ≈3.15 grams isoleucine, ≈5.07 grams of leucine, ≈4.48 grams of tyrosine, ≈2.83 grams of phenylalanine, ≈2.08 grams of histidine, ≈3.63 grams of lysine, ≈3.33 grams of arginine and ≈4.08 grams of proline ((Diender et al., 2011)). 500 mg of chicken meal will be spread over the box ((Barry, 2004)). Every 12 hours, more food should be added ((Barry, 2004)). The temperature will be 30˚ celsius with 60%

Relative Humidity ((Tomberlin et al.,2009)). The moisture level will be at 70% ((Newby, 1997)). The lighting will be 200µ of natural sunlight or from a quartz iodine lamp ((Zhang et al.,2010)). The larvae should be under the light in 12 hour intervals ((Zhang et al.,2010)). The larvae should be in clumped distribution determined by food. A quarter of the box, about 0.25m will be separate from the food. This area will be for the feces of the larvae. The larvae will be treated under these conditions for 22-24 days, until they become pupae ((Diender et al., 2011)). When the larvae start to enter the last instar on days 19-24, decrease the amount of food given to 100 g, and then cut off all food given ((Newby, 1997)). For research purposes, another quarter of the box will be separate from the rest of the sections. This is where 50 of the 500 larvae will be grown, so they can be separated for experiments and research purposes. For measuring, they will be taken out by using tweezers, and measured. Measurement will take place every three days on the same larvae.

Pupal:

The temperature will be 30 degrees celsius with 60% relative humidity ((Tomberlin et al.,2009)). The larvae should have eaten all of the chicken meal at this point, leaving nothing but the dirt. Their will be two pre made holes for the pupal to pupate in. The water hole will still have water from a freshwater river with a pH level of 5 ((Diender et al., 2011)). The lighting will be 200µ of natural sunlight or a quartz iodine lamp ((Zhang et al.,2010)). It should be under the light for 12 hours, and out of the light for 12 hours ((Diender et al., 2011)). In this stage, the pupa will live in the holes and caves. They will still live in the same clear box that is 1m * 1m * 1m ((Tomberlin

et al., 2002)). The dirt will still be 5.5cm, and there will still be ridges. The pupae will be in clumped distribution, determined by the tunnels and holes. The pupa will also be placed near an AC that is on 50% power to add to the dryness. The moisture level should be 70% at this point ((Newby, 1997)). For research purposes, another quarter of the box will be separate from the rest of the sections. This is where 50 of the 500 pupae will be grown, so they can be separated for experiments and research purposes. For measuring, they will be taken out by using tweezers, and measured. Measurement will take place on the same pupae every three days. Pupae will be in this stage for two weeks ((Newby, 1997)).

Adult Protocol:

The temperature should still be at 30 degrees celsius with 60% relative humidity ((Tomberlin et al.,2009)). At this stage, there will be two areas for females and males. The distribution of the adults will be clumped by territory. The females should live in tunnels and crevices with dead organic matter, in this case the same chicken feed as used to feed the larvae chicken feed. This chicken feed should not be replenished as often as during the larval stage. Only once every 2-3 days in amounts of 500 g as the flies are not going to eat it ((Diender et al., 2011)). The males should be near the body of water in the middle ((Sheppard et al.,2002)). A quarter of the box, about 0.25m will be separate from the food. This area will be for the feces of the adult flies. The flies should still live in the clear 1m*1m*1m box, and have 200μ of natural sunlight or from a quartz iodine lamp for 12 hours a day ((Zhang et al.,2010)). The moisture level will be at 70% ((Newby, 1997)). No food will be given at this stage as the flies cannot eat ((Newby, 1997)). At this stage, cardboard trays that are 12.5*12.5*7.5 cm that have been placed in fresh water

obtained from rivers will be placed around the male's section ((Newby, 1997)). Twigs, leaves and other natural matter from the same river should be collected and put around the other territories and in the female oviposition location ((Newby, 1997)). These will act as territories for the females to come and mate with them ((Barry, 2004)). Their will be four of these mating territories, including the middle water spot. After air mating, the female will then return to the crevices. These crevices will have leaves, twigs and chicken meal in which the female will lay its eggs in, and later die ((Gobbi et al., 2013)). After the eggs have been laid, a plastic cup will be used to gather the eggs and the surrounding dirt. For research purposes, another quarter of the box will be separate from the rest of the sections. This is where 50 of the 500 adults will be grown, so they can be separated for experiments and research purposes. For measuring, they will be taken out by using tweezers, and measured. Adults will be measured immediately after death as they are hard to catch alive. Adults will be alive for 5-8 days using this method ((Newby, 1997)).""

Results:

The following protocol was used in a laboratory setting by the author and from the following instructions, there was a significant increase in the amount of Black Soldier Flies that were bred. This protocol allowed the amount of Black Soldier Flies gained from breeding to increase ~36% than if the protocol was not used. This increase is necessary as it allows the model organism of the Black Soldier Flies to be an option and a candidate for laboratory use because of their affordability and usefulness (Marzouk, 2016).

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