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EFFECTS OF FOOD QUALITY ON DAPHNIA AMBIGUA

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EFFECTS OF FOOD QUALITY

ON *DAPHNIA AMBIGUA*

by

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ABSTRACT

EFFECTS OF FOOD QUALITY

ON *DAPHNIA AMBIGUA*

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Environmental changes are common in nature and are responsible for variables such as temperate and food quality. *Daphnia ambigua* are an integral part of aquatic ecosystems, providing food sources for aquatic life and keeping phytoplankton from overgrowing. Our goal was to manipulate food quality in order to observe generational and cross-generational effects on *Daphnia*. Existing clones from cultures in the lab were reared in a common garden setting for three generations in order to reduce extraneous variables. Food quality is a key aspect of development; therefore, it was manipulated in order to determine possible effects on clutch and body size. Results showed that *Daphnia* reared in a low quality food environment produced smaller clutch sizes and were bigger in size compared to those reared in the medium and high quality food treatments. On a broader scale these results demonstrate that food quality has an effect on *Daphnia* traits across generations.

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CHAPTER 1

INTRODUCTION

1.1 Background

Transgenerational epigenetics is a phenomenon that is common amongst many animals and plants in the environment (Walsh, La Pierre, & Post 2014). It occurs when the environment induces phenotypic changes, without DNA sequence modification, that persist for multiple generations. Therefore not only does the organism affect the environment, but also the environment impacts the organisms in a fashion that incorporates both current and future generations. Evidence for these transgenerational effects is rapidly accumulating. Yet, we lack a complete understanding of the factors that induce transgenerational effects as well as the duration of these responses.

1.2 Predator, Prey Interactions and Environmental Effects on *D. ambigua*

The waterflea, *Daphnia ambigua*, is a common feature in freshwater environments (Miller 2000, Herbert & Grewe 1985). From an ecological point of view, *Daphnia* serve as a food source for fish and help in the regulation of phytoplankton (Miller 2000, Carpenter et al. 1987, 1992; Elser et al. 1988). *Daphnia* are also a commonly used study organism because they are easily collected, reared, and manipulated in a laboratory setting.

Daphnia routinely experience significant spatial and temporal variability in resource quality. This variation in resource quality spans food types that differ in morphology, and toxicity. For instance, high-quality green algae is typically abundant during the spring in temperate lakes (Walsh, La Pierre, & Post 2014). Yet summer is typically dominated by low-quality cyanobacteria. Cyanobacteria are considered a low quality food source because it is difficult for the *Daphnia* to consume due to its morphology and the deficiency in certain key nutritional elements (Miller 2000). For instance, cyanobacteria lack polyunsaturated fatty acids and sterols (Miller 2000). Also some of the metabolic byproducts of the cyanobacteria may be harmful to *Daphnia* (Miller 2000). Much research has shown that cyanobacteria negatively influence the fitness of *Daphnia* (Hairston *et al.* 1999; Rohrlack *et al.* 2001; Lurling 2003; Sarnelle, Gustafsson, & Hansson 2010; Walsh, La Pierre, & Post 2014). In turn, *Daphnia* modify several morphological and size-related characters that enhance feeding in the presence of filamentous cyanobacteria (Gliwicz & Lampert 1990; Young et al. 1997; Ghadouani & Pinel-Alloul 2002; Bednarska & Dawidowicz 2007). These changes in the traits of Daphnia, in turn, present the opportunity for *Daphnia* to modify the traits of future generations and to exhibit transgenerational plasticity in response to exposure to cyanobacteria.

1.3 Research Goals

This research explored the transgenerational effects of variation in food quality on life history traits in *Daphnia.* To test this hypothesis, I reared multiple clones of *Daphnia ambigua* from lakes in Connecticut across an increasing gradient in cyanobacteria (*Anabaena*) and assessed this influence on the traits of *Daphnia* across multiple experimental generations. *Anabaena* is a lower quality food source due to its filamentous shape, which makes it difficult for it to be ingested by the *Daphnia.* It is predicted that *Daphnia* that receive the high quality treatment will produce more eggs and will be bigger in size compared to those who receive the lesser quality treatments, and this effect will extend across generations.

1.4 Significance of the Problem

Daphnia play a critical role in the ecosystem, especially in the aquatic food web, because they play a role in the growth of phytoplankton and serve as a food source for fish and other aquatic organism (Miller 2000, Pfrender 2014). Therefore, the effects on *Daphnia* by cyanobacteria may have adverse effects not only on the developmental traits of *Daphnia* but also on the ecosystem as a whole (Miller 2000). An increased understanding of the extent to which the sub-optimal cyanobacteria affect the size, maturation, reproductive success, and population growth of *Daphnia* may be helpful in elucidating the intricate workings and functions of the aquatic ecosystem. This is especially beneficial for areas that have an economy that is dependent on fishing.

Figure 1.1: *Daphnia Ambigua*
(Source: Personal photographs)

Figure 1.2: *Daphnia Ambigua*
(Source: Personal photographs)

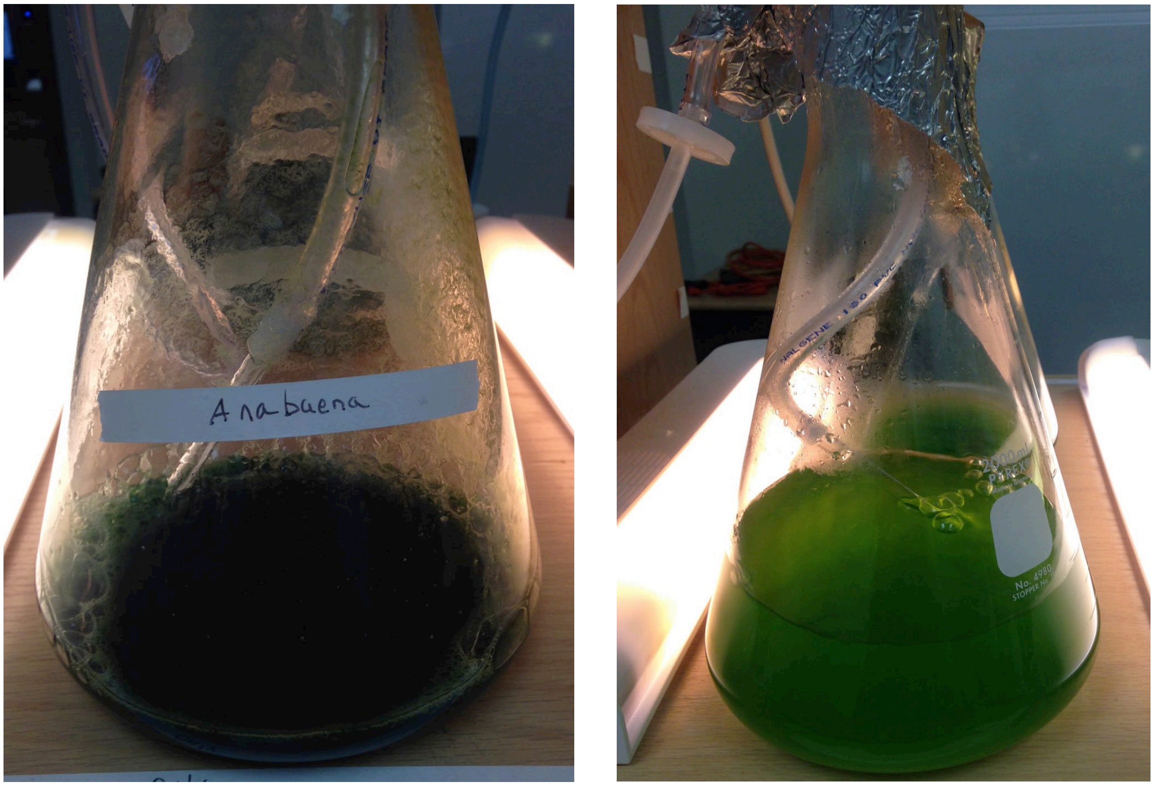

Figure 1.3: Green algae - *Anabaena sp* (left) and *Scenedesmus ob* (right).
(Source: Personal photographs)

CHAPTER 2

METHODOLOGY

2.1 Sample Preparation

The goal of this experiment is to characterize the transgenerational effects of *Anabaena sp* on *Daphnia* life histories. To determine these effects, *Daphnia* was grown in a common laboratory setting for two generations and then exposed to multiple generations of experimental manipulation. Four mature females from existing clones were collected from cultures in the lab and placed individually in 90-ml jars containing COMBO media (Kilham et al. 1998). The clones came from five lakes in Connecticut including Bride, Dodge, Gorton, Long, and Quonnipaug (Post et al. 2008). These individuals were reared under common temperature (18°C) and photoperiod regimes (Photoperiod: 14L:10D). All clones were transferred to fresh media and algae every other day. The second laboratory generation was then established by collecting two newly-born neonates from the second clutch of each clone. The density of individuals was reduced to one individual per container on day 3. These individuals experienced the same conditions (temperature, photoperiod, food quantities) and frequency of food/media replenishment (every day) as the previous generation.

2.2 Experimental Jars and Data Collection

Beginning with the third laboratory reared generation, I measured the effects of cyanobacteria on *Daphnia* traits by rearing multiple clones of *Daphnia* across an increasing gradient in the concentration of *Anabaena*. This experiment ran for two experimental generations (F_1 and F_2). To begin the experiment, I collected nine newly born individuals (<12 hours old) per clone and individually placed them into 90-ml jars containing COMBO media (Kilham et al. 1998). Each individual was randomly assigned to one of three treatments: (1) 100% *Scenedesmus* (green algae) only, (2) 40% *Anabaena sp*/60% *Scenedesmus*, and (3) 80% *Anabaena*/20% *Scenedesmus*.

Beginning on day 3 of the experiment, all *Daphnia* were monitored for the attainment of maturation (defined as the release of the first clutch of offspring into the brood chamber). When an individual matured, a picture was taken for an estimate of size at maturation and the size of the first clutch was recorded. All individuals were subsequently monitored for the production of the second clutch. The transition between the F_0 generation and F_1 generation occurred when the second clutch of offspring was born. Newly born individuals from this clutch were collected and placed into the same treatment as experienced by the parent. The F_1 generation was monitored as stated above and their maturation date and clutch dates and sizes were also recorded using the same procedures as above.

2.3 Data Analysis

Variation in age and size at maturation and clutch size were analyzed using a two-way analysis of variance with generation and resource treatment entered as fixed effects.

CHAPTER 3

RESULTS

3.1 Average Clutch Size

The inclusion of cyanobacteria as a food source significantly ($p<0.05$) influenced reproductive outputs (Fig. 3.1). Overall, *Daphnia* fed 100% *Scenedesmus* produced 25% more offspring than the treatments that were fed *Aanbaena* ($F_{2,124} = 47.51$, $p < 0.001$). The differences among food treatments also varied across generations as I observed a significant food x generation interaction ($F_{2,124} = 15.8$, $p < 0.001$). This because the clutch size of *Daphnia* fed 100% Scenedesmus increased between generation 1 and 2 while those fed Anabaena (40% and 80%) both declined across generations. For the *Daphnia* which received the 40/60 or 80/20 mixture of *Scenedesmus* and *Anabaena sp* the average clutch size declined by 29% and 18% respectively (Fig. 3.1).

Average Clutch Size Across Generation			
Treatment	F_1 Average Clutch Size	F_2 Average Clutch Size	% Change
100 *Scendesmus*	20	25	25
60% *Scendesmus*/40% *Anabaena* sp.	17	12	29
60% *Scendesmus*/40% *Anabaena* sp.	17	14	18

Table 3.1: Average Clutch Size Across Generation

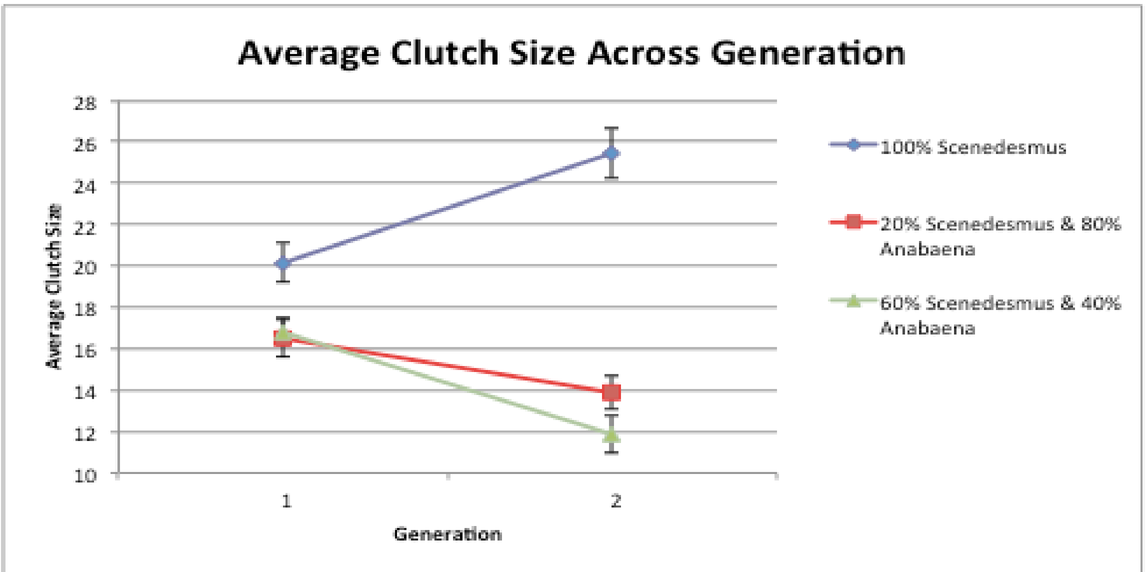

Figure 3.1: Average Clutch Size Across Generations

Tests of Between-Subjects Effects					
Dependent Variable: total					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
food	1768.844	2	884.422	47.508	.000
gen	17.569	1	17.569	.944	.333
food * gen	588.199	2	294.100	15.798	.000
a. R Squared = .458 (Adjusted R Squared = .437)Ta					

Table 3.2: ANOVA Statistical Analysis of Clutch Size

3.2 Average Size of *Daphnia*

The addition of Anabaena in the diet of *Daphnia* did not significantly influence size at maturation ($F_{2,124}$ = 0.64, p = 0.53). The interaction between food treatment and generation was marginally non-significant (Fig. 3.2) ($F_{2,124}$ = 2.4, p =0.095). This is because Daphnia reared on a diet that consisted of 80% Anabaena increased size at maturation between generation 1 and 2 (a ~6% increase) while the other two treatments exhibited modest changes in size between the two experimental generations (Fig. 3.2).

Average Feret Size Across Generations			
Treatment	F_1 Average Feret Size (mm)	F_2 Average Feret Size (mm)	% Change
100 *Scendesmus*	4.04	4.01	.74
60% *Scendesmus*/40% *Anabaena* sp.	4.05	3.96	2.22
60% *Scendesmus*/40% *Anabaena* sp.	3.88	4.10	5.67

Table 3.3 Average Length at Maturation

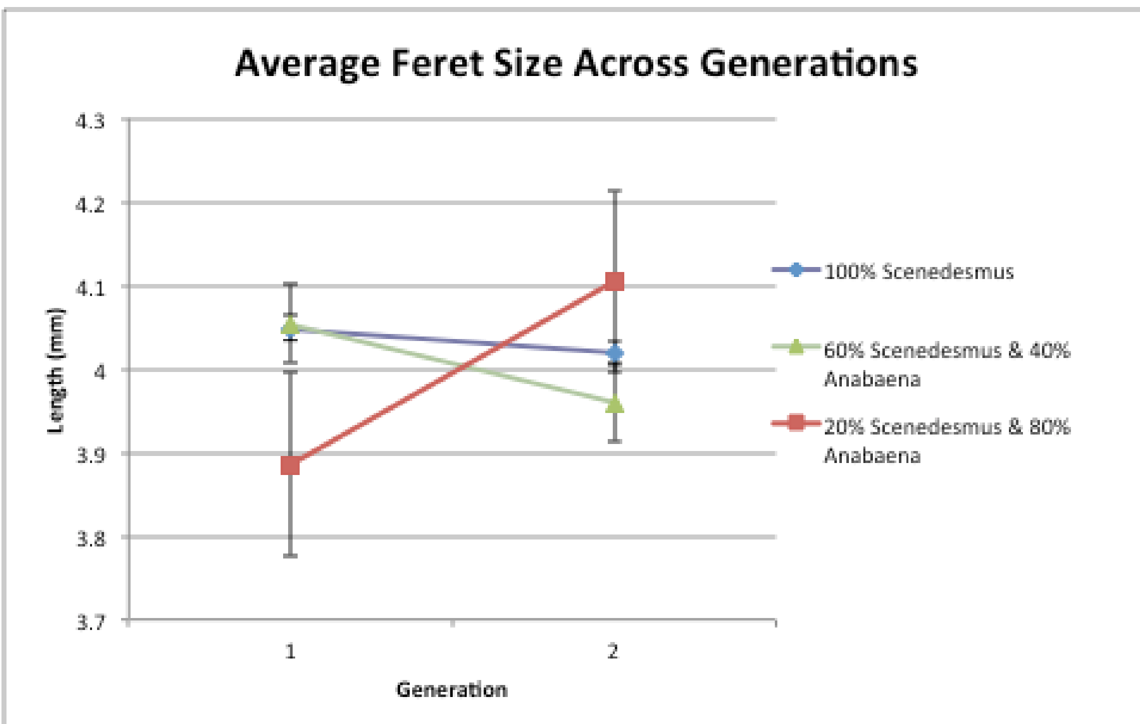

Figure 3.2: Average Length at Maturation

Tests of Between-Subjects Effects					
Dependent Variable: area					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
food	1.218	2	.609	.641	.528
gen	.086	1	.086	.091	.764
food * gen	4.555	2	2.277	2.398	.095
a. R Squared = .061 (Adjusted R Squared = .023)					

Table 3.4: ANOVA Statistical Analysis of Length

CHAPTER 4

DISCUSSION

4.1 Data Analysis

Determination of *Daphnia* plasticity is an important concept because it can serve as an indicator of changing environments, and this study found that manipulation of food quality can also reveal information about transgenerational plasticity. It was hypothesized that *Daphnia* that received the higher quality treatment would produce bigger clutches and would be bigger in size compared to those who received the sub-optimal food treatments. This study shows that *Daphnia* reared on high quality food did produce more offspring, but there was no corresponding influence on size (Fig. 3.1-2). This study also showed that variation in resource quality influenced the expression of life history traits across generations. *Daphnia* reared on high quality diets increased the size of their clutches between generations while the opposite response was observed when *Daphnia* was reared on sub-optimal cyanobacteria (Fig. 3.1). There was some evidence that *Daphnia* may adaptively respond to the presence of cyanobacteria by increasing their size at maturation in the following generation (Fig. 3.2). Such a response may improve the handling or ingestion of this low quality food source. However, the result present in this paper was non-significant and requires further investigation.

4.2 Significance of Concept

The concept of transgenerational plasticity applies not only to *Daphnia*, but also to humans. An example of this is can be seen in fetuses in the womb that are exposed to stress. When the body is stressed the sympathetic nervous system takes over and produces a flight or fight response. In this state the body releases stress hormones such as cortisol and epinephrine to get it ready for future stressful situations. This system serves to keep the body aware of dangerous situations; however, chronic cortisol and epinephrine release is harmful not only to the individual but also to the fetus. There have been past studies which indicate that chronic stress can lead to premature births and low birth weights (WebMD, 2015). Another example of this can be seen from offspring produced during the Dutch Famine of 1944 (Lumey et al. 2007). Children born during this time of hardship usually weighed less than those produced prior to or after the famine. The effects of transgenerational effect persisted for two generations before returning the birth weight returned to average.

4.3 Conclusion and Areas of Future Study

Future studies can compare the effects of food treatments on transgenerational phenotypes of other species of *Daphnia,* including but not limited to *D. pulex* and *D.* magna, to see if the transgenerational effects of resource quality are consistent across species. Both *D. pulex* and *D. magna* are substantially bigger than *D. ambigua.* Such experiments may therefore reveal novel insights into the transgenerational effects of resource quality. The experimental organism can also be used with other variables such as temperature, water quality and predators to further define the transgenerational effects of environmental stimuli.

REFERENCES

Bashey, F. (2006). Cross-generational environmental effects and the evolution of offspring size in the Trinidadian guppy Poecilia reticulata. *Evolution*, *60*(2): 348-361.

Carpenter, S. R. et al. (1987). Regulation of lake primary productivity by food web structure. *Ecology* 68: 1863–1876.

Hebert P.D.N. and Grewe P.M. (1985). Chaoborus-Induced Shifts in the Morphology of *Dapnhia ambigua*. *American Society of Limnology and Oceanography* 30: 1291-1297.

Kilham, S.S., Kreeger, D.A., Lynn, S.G., Goulden C.E., Herrera, L. (1998). COMBO: a defined freshwater culture medium for algae and zooplankton. *Hydrobiologia,* 377: 147-159.

Lumey LH., Steing AD, Kahn HS, van der Pal-de-Bruin KM, Blauw GJ, Zybert PA, Susser ES. (2007 May) Cohort Profile: The Dutch Hunger Winter Families Study. International Journal of Epidemiology. [date accessed: 2015 March 14]; 36(6): 1196-1204. doi: 10.1093/ije/dym126

Miller, C. 2000. "Dapnhia pulex" (On-line), Animal Diversity Web. Accessed November 07, 2014 at http://animaldiversity.ummz.umich.edu/accounts/Dapnhia_pulex/

Pfrender ME. Dapnhia [Internet]. [Cited 2014 Nov 7] . Available from: http://www.nih.gov/science/models/dapnhia/

Riessen, H.P. (1999). Predator-induced life history shifts in *Dapnhia*: a synthesis of studies using meta-analysis. *Canadian Journal of Fisheries and Aquatic Sciences* 56(12): 2487-2494.

Spitze K. (1992). Predator-Mediated Plasticity of Prey Life History and Morphology*: Chaoborus americanus* Predation on *Dapnhia pulex*. *The American Naturalist* 139: 229-247.

Walsh M.R., La Pierre K.J., Post D.M. (2014) Phytoplankton composition modifies predator-driven life history evolution in *Dapnhia*. Evol Ecol. 28: 397-411.

WebMD. (2015). [accessed 2015 April 23]. http://www.webmd.com/baby/features/fetal-stress?page=3

BIOGRAPHICAL INFORMATION

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