Population Differences in Fecundity Components in the Hermaphroditic Freshwater Snail *Planorbella trivolvis*

Author(s): Cynthia G. Norton, Angela F. Johnson and Betsy M. Nelson
Source: American Malacological Bulletin, 36(1):23-30.
Published By: American Malacological Society
https://doi.org/10.4003/006.036.0109
URL: http://www.bioone.org/doi/full/10.4003/006.036.0109

Amer. Malac. Bull. **36**(1): 23–30 (2018)

Population differences in fecundity components in the hermaphroditic freshwater snail *Planorbella trivolvis*

Cynthia G. Norton[1], Angela F. Johnson[1], and Betsy M. Nelson[1]

[1]Department of Biology, St. Catherine University, 2004 Randolph Ave. St. Paul, Minnesota, 55105, U.S.A., cgnorton@stkate.edu

Abstract: Fecundity in outcrossing species can be influenced by both maternal and paternal parents. To tease out these influences, we observed egg production in two populations of the hermaphroditic freshwater snail, *Planorbella trivolvis* (Say 1817). We carried out both intra- and inter-population matings between individuals from an inbred albino laboratory strain and individuals one generation removed from a natural population, and measured egg production for two 3-week periods – immediately after mating and 11 weeks later. In the female role, individuals from the albino laboratory population produced significantly more egg masses than the wild derived snails, regardless of whether they were mated to a partner of wild or laboratory origin, indicating that egg mass production is controlled by the maternal parent. Conversely, regardless of their own origin (laboratory or wild), snails mated to individuals from the wild derived population laid more eggs in each egg mass than those fertilized by sperm from the lab population. These results indicate that the number of egg masses a snail lays is determined at least in part by a maternal contribution, and that the number of eggs deposited in each mass may be influenced by the origin of the sperm donor. In addition, we confirmed a short-term trade-off between growth and reproduction as differences in egg production among mating types were mirrored by differences in body size. Further investigation into the nature of these differences will lead to better understanding of the reproductive biology of these hermaphrodite mollusks.

Key words: egg mass production, laboratory and wild populations, albinism

Understanding factors that impact reproductive fitness is key to understanding the evolution of reproductive strategies. Appreciating the full realm of these influences involves the study not only of organisms in which the sexes are separate (gonochorists), but also in hermaphrodites, organisms which reproduce as both male and female – either sequentially or simultaneously (Anthes *et al.* 2010, Schärer *et al.* 2014). Our laboratory uses the freshwater hermaphroditic snail, *Planorbella trivolvis* (Say 1817 Hygrophila: Planorbidae, formerly known as *Helisoma trivolvis*), as a model system for addressing key questions about reproduction and mating strategies in hermaphrodites to answer recent calls for empirical studies to test current theories (Nakadera and Koene 2013, Schärer *et al.* 2014). *P. trivolvis* is a hermaphroditic freshwater Pulmonate snail widely distributed across the US and Canada (Johnson *et al.* 2013). Individuals may mate as male, delivering sperm to a partner, or as female, producing eggs, but often mate reciprocally, acting simultaneously as male and female (Abdel-Malek 1952 and personal observations). Following mating, eggs are laid approximately daily in masses of about 20 individual eggs for more than 16 weeks (Norton and Newman 2016). Although most hermaphroditic freshwater snails will produce self-fertilized eggs when isolated, *P. trivolvis* rarely produce selfed offspring in the laboratory (Crabb 1927, Paraense and Correa 1988, Escobar *et al.* 2011, Norton and Newman 2016, Norton *et. al.* 2018).

Fecundity in outcrossing species can be influenced by both maternal and paternal parents, the sources of egg and sperm. In hermaphrodites, these individuals are typically referred to as the sperm recipient (here, egg layer or producer) and the sperm donor. To tease out these influences, we observed egg production in two populations of snails (one long term laboratory strain and one from wild collections) and crosses between the two populations. The total number of eggs produced by a snail is influenced by both the number of egg masses laid and the number of eggs in each mass; our goal was to determine whether the source of the egg producer and/or sperm donor influenced these two fecundity components. Since these snails store sperm, we were also interested in whether populations might differ in sperm transfer, longevity, and/or storage capacity. We therefore assessed egg production components both immediately after mating and 10 weeks later as allosperm (from a partner) was being depleted. Finally, population differences in fecundity may influence growth due to trade-offs between somatic and reproductive efforts (Koene and terMaat 2004, Norton and Newman 2016), so we measured body size as well as egg production.

MATERIALS AND METHODS

Study populations

Laboratory *Planorbella trivolvis* were sampled from an albino population originally derived from two non-pigmented individuals found in a laboratory stock in 2002. Wild snails

were the progeny of individuals collected from a pond at the entrance to Charles Towne Landing State Park (Charleston, NC 32.8068°N; 79.9902°W). The test snails came from eggs laid by nine individuals from each population within a one week period as part of another study to investigate the genetic basis of albinism (Norton *et. al.* 2018).

Matings

We isolated 54 snails from each population (6 from each adult) at 10 weeks of age in 296 ml plastic cups filled with dechlorinated tap water. We fed them boiled romaine lettuce (approx. 2x2 cm, an amount typically eaten in 3–4 days by adult snails housed alone in the lab) and changed water twice per week. Snails were kept in the laboratory at room temperature (~22 °C) with ambient light on a rolling cart with multiple shelves. Matings were set up when snails were approximately 126 days old, well after sexual maturity (104.3 ± 9.7 d = 14.9 weeks; Escobar *et al.* 2011). Snails were paired randomly with a partner in a single cup for one week, and then removed to individual cups one week later. We set up 18 pairs of each type: Laboratory x Laboratory, Wild x Wild, and Laboratory x Wild. The within population matings (N = 36 snails in each), allowed us to characterize egg production in the two populations. Since both parents produce eggs as well as contribute sperm to their partner, the crosses between populations also resulted in 36 focal individuals, half of which (N = 18) were albinos from the laboratory population mated to a pigmented wild snail and the other half (N = 18) were their pigmented wild partners. Because *P. trivolvis* rarely, if ever, self-fertilize (Norton and Newman 2016, Norton *et. al.* 2018) we can safely assume that sperm always originates from the partner (allosperm).

Egg production and body size

To determine if there were differences in fecundity components between the laboratory-derived snails and those of wild origin, we counted all egg masses laid and the number of eggs in each mass for the first three weeks after mating for all individuals. We determined the number of egg masses, the average number of eggs per mass and overall number of eggs produced by each snail during these three weeks. We then calculated the average number of egg masses laid, average number of eggs per mass, and the average overall number of eggs produced for snails in each of the four population cross combinations (Laboratory x Laboratory, Laboratory x Wild, Wild x Laboratory and Wild x Wild). Similarly, we counted eggs and egg masses for three weeks at 11-13 weeks post-mating, once allosperm was becoming depleted. We also measured shell diameter at the time of mating and 11 weeks later using handheld digital calipers, a highly reliable measurement of body size (Norton *et al.* 2008). Because body size influences egg production in this species (Norton and Bronson 2006), we used body size as a covariate in population comparisons of egg production at both 1 and 11 weeks post-mating. We initially compared egg production between the two original populations using a one-way ANOVA. To determine maternal and paternal contributions to these egg production parameters, we compared egg production among the four mating combinations using a two-way ANOVA examining the main effects of egg source and sperm source and any interaction. All analyses were done using IBM SPSS Version 22. Any individual that did not lay eggs in the first three-week observation period was omitted from these analyses, so final sample sizes reflect these omissions.

RESULTS

Egg production

Almost all snails housed with a partner for one week laid eggs (Table 1). Although total egg production during the first three weeks after mating did not differ significantly between the laboratory (Laboratory x Laboratory) and wild (Wild x Wild) populations (Figure 1A; ANOVA: $F_{[1,60]} = 1.91$, $P = 0.17$), individuals from the albino laboratory population laid significantly more egg masses than those from the pigmented wild population (Figure 1B; one-way ANOVA: $F_{[1,60]} = 13.38$, $P = 0.001$) but significantly fewer eggs in each mass (Figure 1C; one-way ANOVA: $F_{[1,60]} = 17.50$, $P < 0.001$).

To explore these differences further, we analyzed egg production differences among all four combinations, the within-population matings (Laboratory x Laboratory and Wild x

Table 1. Reproductive activity of snails mated within and between populations.

Cross	Number of Snails Mated	Number Laying Eggs 1 week post-mating (%)	Number Laying Eggs 11 weeks post-mating (%)
Laboratory x Laboratory	36	30 (83.3)	8 (22.2)
Laboratory x Wild	18	17 (94.4)	16 (88.9)
Wild x Laboratory	18	13 (72.2)	7 (38.9)
Wild x Wild	36	33 (91.2)	29 (80.1)
All Snails	108	93 (86.1)	60 (55.6)

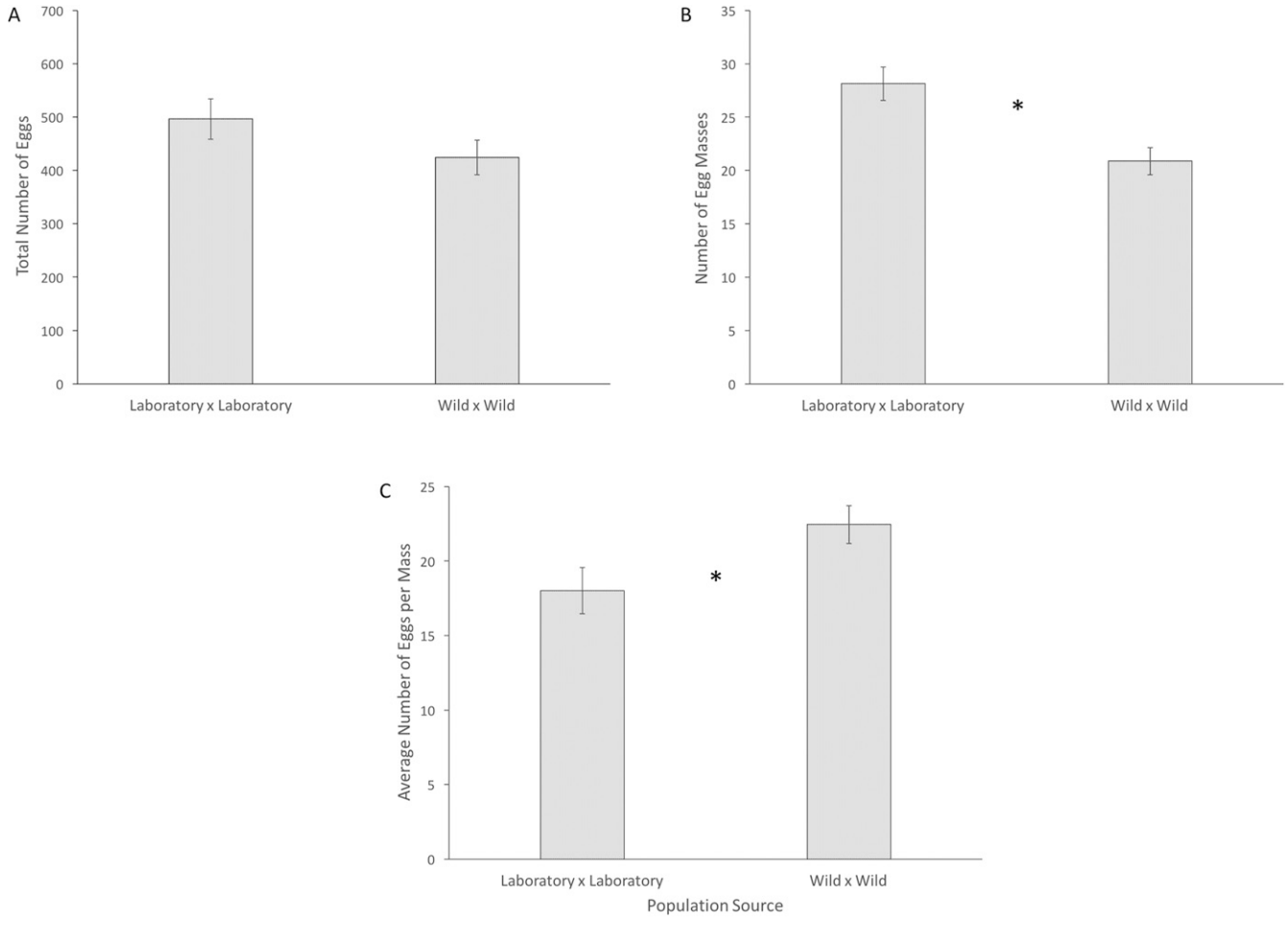

Figure 1. Egg production in snails from two sources, an albino laboratory population and a pigmented wild population 1-3 weeks post-mating. A, Mean number of eggs produced by snails from each population during a 3-week period immediately following mating. B, Average number of egg masses laid. C, Average number of eggs in each egg mass. An asterisk indicates significant difference between means. Error bars represent ± SE.

Wild), and between-population crosses (Laboratory x Wild and Wild x Laboratory). Analyses are summarized in Table 2. During the first three weeks after mating, total egg production was influenced by the origin of both the egg producer and sperm donor (Figure 2A; two-way ANOVA, egg source: $F_{[1,88]} = 16.71$, $P < 0.001$; two-way ANOVA, sperm source: $F_{[1,88]} = 5.79$, $P = 0.018$). There was a significant main effect of the egg layer's origin on the number of egg masses laid; snails from the albino laboratory population, regardless of the origin of their mate (the sperm donor), laid significantly more egg masses than pigmented snails from the population of wild origins (Figure 2B; two-way ANOVA: $F_{[1,88]} = 28.78$, $P < 0.001$). Conversely, there was a significant effect of sperm donor origin on the average number of eggs per egg mass. Those individuals mated to a sperm donor from the wild population laid significantly more eggs per egg mass than those mated to a snail from the laboratory population, again regardless of the origin of the egg producer (Figure 2C; two-way ANOVA: $F_{[1,88]} = 17.15$, $P < 0.001$).

By 11 weeks post-mating, fewer of the snails were producing eggs, particularly those mated to a sperm donor from the laboratory population (Table 1). Those still laying eggs were producing fewer egg masses (14.1 ± 1.05 vs 25.2 ± .91; paired t-test: $t_{[59]} = 10.02$, $P < .001$) and fewer eggs per mass (12.7 ± 1.22 vs 21.6 ± .43; paired t-test: $t_{[59]} = 7.56$, $P < .001$) than they were the first three weeks after a mating opportunity. Although there were still differences among treatments in the total number of eggs produced (Figure 3A), these differences are explained solely by the source of the sperm donor (two-way ANOVA: $F_{[1,88]} = 8.81$, $P = 0.004$). As was the case immediately after mating, individuals mated to a sperm donor from the wild population laid significantly more eggs

Table 2. ANOVA tables for 2-way analysis of egg production at 1 and 11 weeks post-mating for snails mated within and between populations.

		1 Week Post-mating						11 Weeks Post-mating					
		Total Number of Eggs		Number of Egg Masses		Average Number of Eggs per Mass		Total Number of Eggs		Number of Egg Masses		Average Number of Eggs per Mass	
Source of Variation	**df**	**F**	**p**	**F**	**p**	**F**	**p**	**F**	**p**	**F**	**p**	**F**	**p**
Egg Source (Laboratory or Wild)	1, 88	16.71	<0.001	28.78	<0.001	1.36	0.25	1.86	0.18	0.65	0.42	0.01	0.92
Sperm Source (Laboratory or Wild)	1, 88	5.79	0.018	0.75	0.39	17.15	<0.001	8.81	0.004	9.04	0.003	11.58	0.001
Interaction (Egg x Sperm Source)	1, 88	3.19	0.077	1.98	0.16	0.66	0.42	0.40	0.53	5.58	0.02	0.17	0.68

per mass than those mated to a sperm donor from the laboratory population (Figure 3C; two-way ANOVA: $F_{[1,88]} = 11.58$, $P = 0.001$). However, snails mated to wild sperm donors also laid significantly more egg masses than those mated to snails from the laboratory population (Figure 3B; two-way ANOVA: $F_{[1,88]} = 9.04$, $P = 0.003$). In addition there was a significant interaction between the source of sperm donor and recipient (two-way ANOVA: $F_{[1,88]} = 5.58$, $P = 0.02$). Laboratory recipients produced more egg masses when mated to a wild sperm donor than to a laboratory donor.

Body size and egg production

This late-term effect of sperm donor on egg production was also reflected in body size differences (Figure 4). There were no significant differences in shell diameter among snails in the four mating types when they were paired (Figure 4A; one-way ANOVA: $F_{[3,89]} = 1.71$, $p = 0.18$) and no significant relationship between body size and egg production at this time ($R^2 = 0.002$; $F_{[1,91]} = 2.19$, $p = 0.64$). However, 11 weeks later there was a significant relationship between shell diameter and total three-week egg production ($R^2 = 0.355$; $F_{[1,91]} = 50.05$, $p < 0.001$).

There was a significant effect of sperm donor on shell diameter at 11 weeks post-mating (Figure 4B; ANOVA $F_{[1,89]} = 26.56$, $p < 0.001$), but no effect of an individual's own origin (ANOVA $F_{[1,89]} = 0.18$, $p = 0.67$). Those individuals mated to a sperm donor from the wild population (and thus producing eggs for a longer time) were significantly smaller than those mated to a partner from the laboratory population (many of which had ceased laying eggs, or were laying fewer than those with wild-origin partners). Those snails mated to partners from the wild population also grew significantly less than those mated to snails from the laboratory population (ANOVA $F_{[1,89]} = 25.70$, $p < 0.001$). In both cases, there was a significant interaction term (ANOVA $F_{[1,89]} = 11.92$, $p = 0.001$ for shell diameter 11 weeks after mating and ANOVA $F_{[1,89]} = 5.70$, $p = 0.02$ for growth over those 11 weeks), indicating that the effect of the sperm donor was dependent on the source population of the individual (laboratory or wild).

DISCUSSION

Our data demonstrate population differences in fecundity components that implicate influences of the egg layer (sperm recipient) on rates of egg mass production and of sperm donor on the number of eggs laid in each mass. In addition, they confirm our earlier reports of a trade-off between growth and reproduction.

Egg production

Differences in egg production between two populations and reciprocal between-population crosses have allowed us

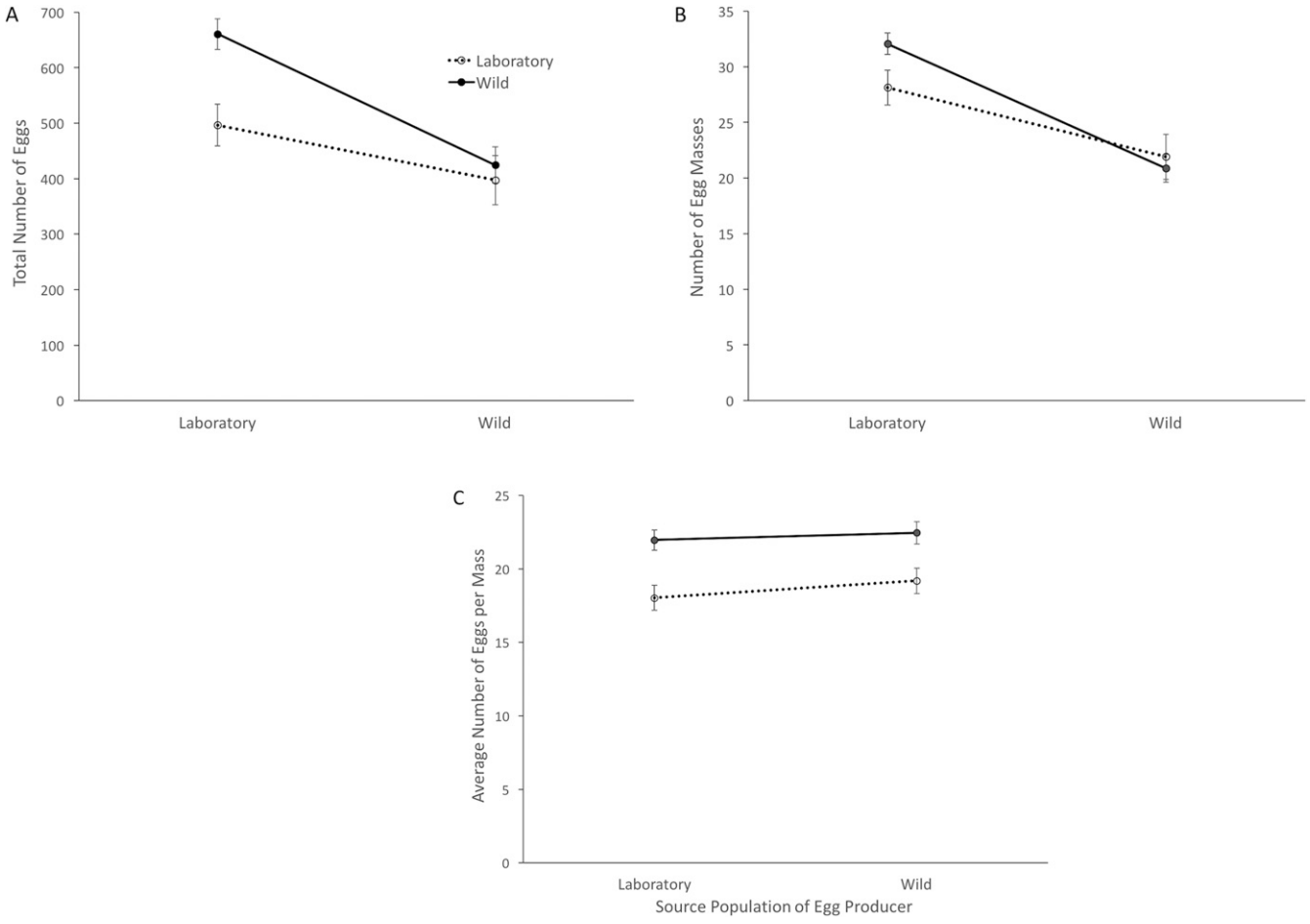

Figure 2. Egg production from within and between population crosses 1-3 weeks post-mating. Source of the egg producer (sperm recipient) is indicated on the x-axis and each line indicates the source of the sperm donor; laboratory sperm donors are indicated by open circles and connected by dotted lines, wild sperm donors are indicated by closed circles and solid lines. A, Mean number of eggs produced by snails during a 3-week period immediately following mating. B, Average number of egg masses laid. C, Average number of eggs in each egg mass. Error bars represent ± SE.

to dissect influences of the sperm recipient and sperm donor on overall egg production. During the first three weeks after mating, the number of egg masses laid by snails is influenced by the origin of the maternal partner, the individual providing the eggs as well as any accessory fluids and packaging (Geraerts and Joosse 1984). Individuals from the albino laboratory population laid more egg masses than those from the pigmented wild origin population, independent of the source of the sperm donor. Conversely, the number of eggs per egg mass appears to be determined at least in part by the origin of the sperm donor. Individuals mated to a partner from the wild origin pigmented population sequestered more eggs in each egg mass than those mated to an individual from the laboratory albino population, again independent of their own origin. It makes sense that the regularity with which egg masses are produced would be determined by the qualities of the individual producing the eggs and accessory proteins and carbohydrates composing the egg jelly (Geraerts and Joosse 1984). It is also logical that the number of eggs in each mass would be determined by the availability of sperm. This observation is supported by observations of egg production 11 weeks after mating.

At 11 weeks post-mating, differences in egg production between snails originating from the laboratory and wild derived populations are explained solely by the origin of the sperm donor. Individuals mated to a pigmented sperm donor from the wild population laid more egg masses with greater numbers of eggs in each mass than individuals mated to a sperm donor from the albino laboratory population. At this point in time differences in female reproductive output may be limited not by their own physiology but by sperm availability. These differences could result from differences among

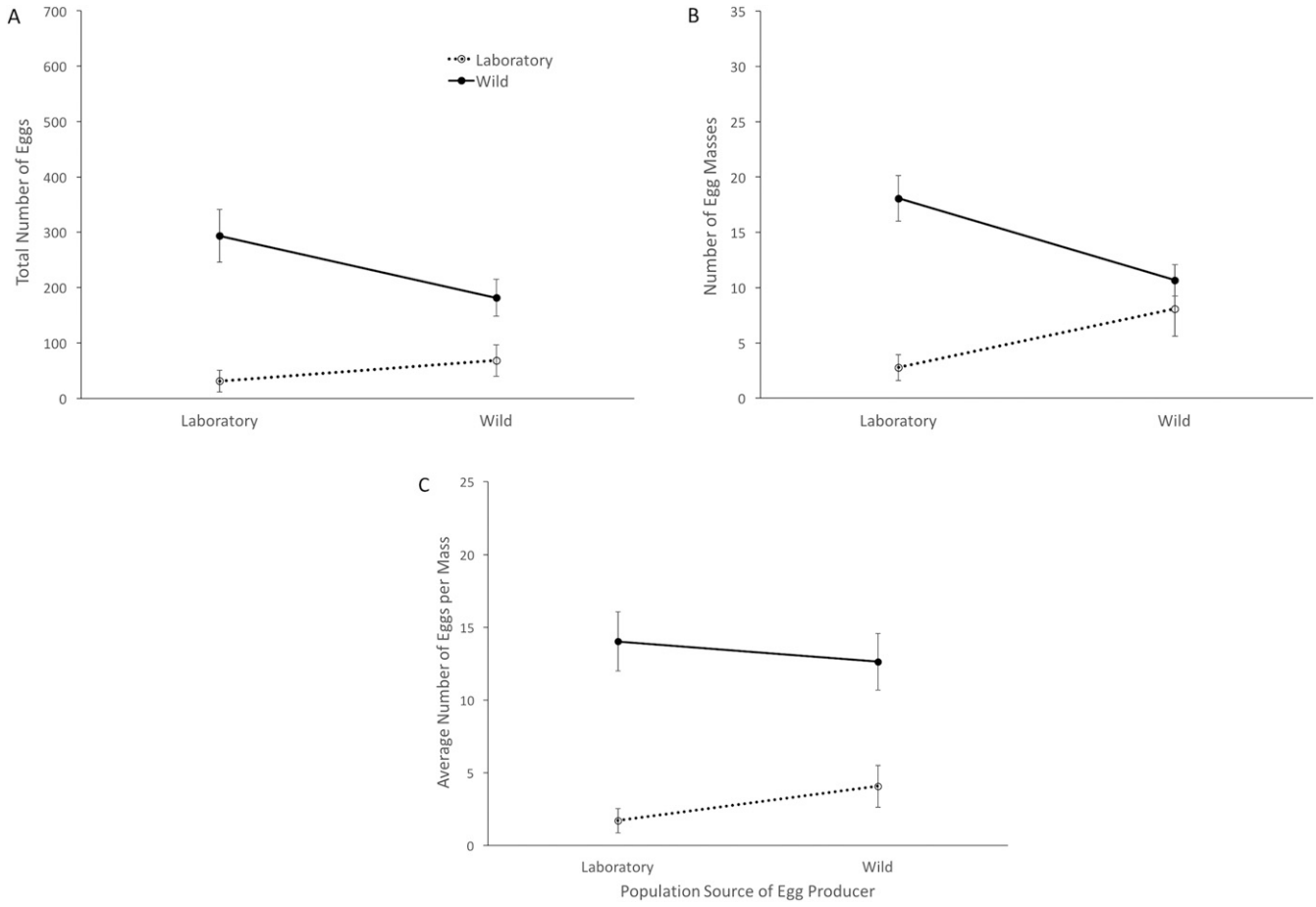

Figure 3. Egg production from within and between population crosses 11-13 weeks post-mating. Source of the egg producer (sperm recipient) is indicated on the x-axis and each line indicates the source of the sperm donor; laboratory sperm donors are indicated by open circles and connected by dotted lines, wild sperm donors are indicated by closed circles and solid lines. A, Mean number of eggs produced by snails in each group during a 3-week period immediately following mating. B, Average number of egg masses laid. C, Average number of eggs in each egg mass. Error bars represent ± SE.

source populations in the number of sperm transferred, possibly as a result of different mating durations, or the longevity of sperm. A sperm donor effect on egg production has not been reported before, nor have differences between pigmented and albino individuals in the number of sperm transferred or longevity of stored sperm.

Planorbella trivolvis store sperm an average of 16 weeks (Norton and Newman 2016), but there is variation among individuals in the duration of sperm storage which could be due to differences in sperm longevity, the amount of sperm transferred, and /or the capacity of sperm storage organs. Interactions between sperm longevity and storage capacity may explain the significant interaction term between egg and sperm source for egg masses produced from 11–13 weeks after mating. Further studies on the relationships among copulation duration, the amount of sperm transferred, sperm storage, and egg production will be necessary to sort out what specific factors are responsible for the differences we have observed.

Although the most obvious difference between our two source populations is the difference in pigmentation, we are *not* proposing that the differences in egg production documented here are solely caused by different alleles at the albino locus. It is possible that some of these differences are either pleiotropic effects of albinism or related to the effects of closely linked genes that might influence sperm or egg production. This was likely the case for fecundity differences between albino and pigmented *Biomphalaria glabrata* (Say 1818) reported by Vianey-Liaud (1989) since both strains originated from the same population. Our finding of more egg masses laid by laboratory-derived albino snails is consistent with their observation that isolated, pure-breeding albino individuals produced more egg masses than pigmented

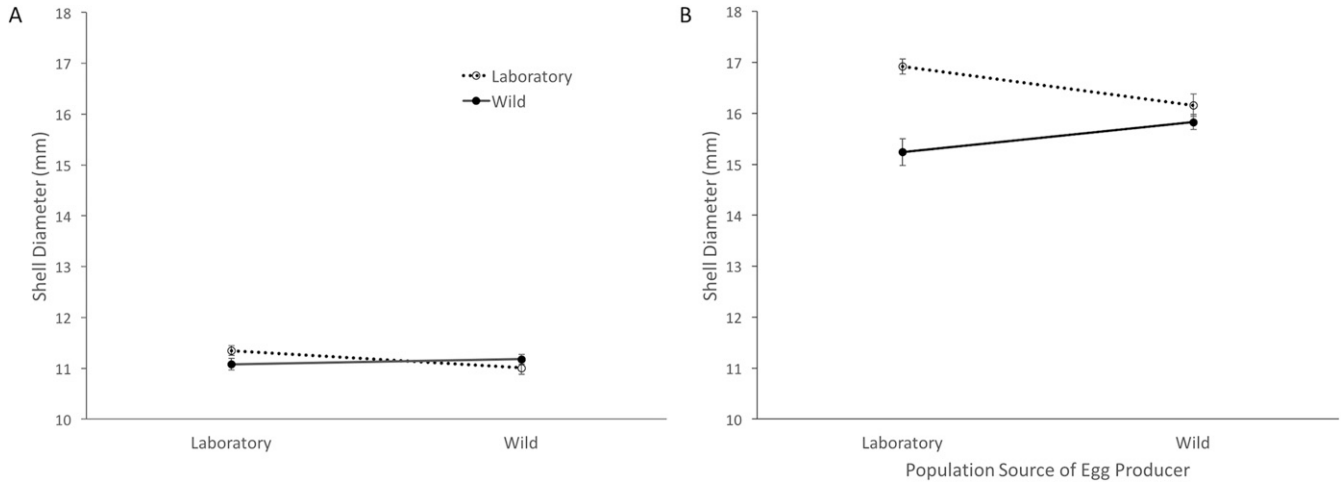

Figure 4. Body size of snails at mating and 11 weeks later. Source of the egg producer (sperm recipient) is indicated on the x-axis and each line indicates the source of the sperm donor; laboratory sperm donors are indicated by open circles and connected by dotted lines, wild sperm donors are indicated by closed circles and solid lines. A, Shell diameter of snails at the time of mating. B, Shell diameter of snails 11 weeks post-mating. Error bars represent ± SE.

individuals, even though they were smaller in size at first reproduction. However, in contrast to our findings, *B. glabrata* albinos also laid more eggs in each mass than did pigmented snails. Vianey-Liaud *et al.* (1996) report that sperm from pure-breeding pigmented individuals is longer than that from albinos, which may be related to velocity and thus provide a competitive advantage. Sperm competition might explain relative numbers of albino and pigmented progeny, but not overall numbers of eggs packaged into each mass unless the sperm differences are not linked directly to the allele they carry, but the genetic make-up of the individual in which they are produced.

In the present study, it seems more likely that the observed differences in egg production are simply differences among source populations. The albino laboratory population originated from snails maintained in the laboratory for many years, and the pigmented snails were one generation removed from the wild, so these differences may result from forces of selection in the laboratory setting that are different from those in the field. Studier *et al.* (1975) found that snails from a laboratory-reared strain of *Helisoma trivolvis* (Say 1817, now *Planorbella trivolvis*, also albino) allocated almost twice as much of their daily energy budget to egg production than did snails from a wild population – but their low assimilation efficiency relative to wild snails may account for their rarity in natural populations. Falade and Otarigno (2013) demonstrated that as wild *Biomphalaria pfeifferi* (Kraus 1848) acclimated to a laboratory setting the number of eggs per snail, number of egg masses per snail and number of eggs per mass all increased. It is also possible that the differences we observed among populations from different original sources represent natural variation in egg production parameters. Browne (1978) found substantial differences among four lakes in upstate New York in female fecundity of the freshwater prosobranch *Viviparus georgianus* (Lea 1834), but no other studies have addressed natural variation in egg production in freshwater molluscan populations. We now have albino F_2 individuals in the laboratory that share approximately half their genome with the wild caught populations, so it may be possible to sort out population differences from direct or pleiotropic effects of the albino locus.

Body size and egg production

The differences in egg production among snails with different origins and mating partners also allowed us to confirm earlier observations of trade-offs between growth and reproduction (Norton and Bronson 2006, Norton and Newman 2016). When snails were paired (at 18 weeks of age) there were no differences among snails designated for the four mating types, and thus the two original populations, in body size. However, 11 weeks later, there was a significant effect of the source of the mating partner on both body size and growth. Snails mated to a partner from the pigmented wild population were significantly smaller and grew less than those mated to a laboratory population albino. Most snails mated to a pigmented sperm donor (over 80% of those in the Laboratory x Wild and Wild x Wild crosses) continued to lay eggs at 29 weeks, whereas fewer than 40% of snails mated to an albino partner from the lab population (Laboratory x Laboratory and Wild x Laboratory groups) were laying eggs at this time. Even among those snails that were laying eggs, those with sperm donors from the laboratory albino population were laying

fewer egg masses and fewer eggs per mass, and thus fewer overall eggs, than those with pigmented wild-origin partners. Those snails laying fewer eggs were thus channeling fewer resources into reproduction and were able to grow to larger sizes as a consequence, confirming a short-term trade-off between growth and reproduction. This pattern is similar to that we observed in snails in which we controlled mating in previous experiments; snails afforded mating opportunities and laying eggs grew significantly less than those who were isolated and producing almost no eggs (Norton and Newman 2016).

In summary, these data provide insights into maternal and paternal influences on egg production in the outcrossing hermaphroditic snail, *P. trivolvis*. These factors may impact not only traditional life history parameters such as when, how often and how much energy to allocate to reproduction (Roff 1992, Stearns, 1992), but also the hermaphrodite-specific challenges of gender role and sex allocation (how much energy an organism devotes to male vs female function, Charnov 1982). Further investigation into the nature of these influences will lead to better understanding of the reproductive biology of these hermaphrodite mollusks.

ACKNOWLEDGEMENTS

C. Norton was supported by the Endowed Professorship in the Sciences and A. Johnson and B Nelson were funded by a 3M Faculty/Student Collaborative Grant through St. Catherine University, St. Paul, Minnesota. We thank Rob Dillon for providing the wild-collected snails.

LITERATURE CITED

Abdel-Malek, E. T. 1952. The preputial organ of snails in the genus *Helisoma* (Gastropoda: Pulmonata). *American Midland Naturalist* **48**: 94–102.

Anthes, N., P. David, J. Auld, J. Hoffer, P. Jarne, J. Koene, H. Kokko, M. C. Lorenzi, B. Pélissié, D. Sprenger, A Staikou, L. Schärer. 2010. Bateman gradients in hermaphrodites: An extended approach to quantify sexual selection. *American Naturalist* **176**: 249–263.

Browne, R. A. 1978. Growth, mortality, fecundity, biomass and productivity of four lake populations of the prosobranch snail *Viviparus georgianus*. *Ecology* **59**: 742-750.

Charnov, E. L. 1982. *The Theory of Sex Allocation*. Princeton University Press: Princeton, NJ.

Crabb, E. D. 1927. The fertilization process in the snail, *Lymnaea stagnalis appressa* Say. *Biological Bulletin* **53**: 67–108.

Escobar, J. S., J. R. Auld, A. C. Correa, J. M. Alonso, Y. K. Bony, M. Coutellec, J. M. Koene, J. Pointier, P. Jarne, P. David. 2011. Patterns of mating-system evolution in hermaphroditic animals: correlations among selfing rate, inbreeding depressions, and the timing of reproduction. *Evolution* **65**: 1233–1253.

Falade, M. O., and B. Otarigho. 2013. Survival potential, fecundity and fertility of Biomphalaria pfefferi (Krauss, 1848) during acclimatization in the laboratory. *Zoology and Ecology* **23**: 157-161.

Geraerts, P. M. and J. Joosse. 1984. Freshwater snails (Basommatophora) *In*: A. S. Tompa, N. H. Verdonk, J. A. M. van der Biggelaar, eds. *The Mollusca ,Vol. 7: Reproduction*, 1st Edition. Academic Press: New York. Pp 141–207.

Johnson, P. D., A. E. Bogan, K. M. Brown, N. M. Burkhead, J. R. Cordeiro, J. T. Garner, P. D. Hartfield, D. A. W. Lepitzki, G. L. Mackie, E. Pip, T. A. Tarpley, J. S. Tiemann, N. V. Whelan, E. Strong. 2013. Conservation status of freshwater gastropods of Canada and the United States. *Fisheries* **38**: 247–282.

Koene, J. M., and A. ter Maat. 2004. Energy budgets in the simultaneously hermaphroditic pond snail, *Lymnaea stagnalis*: a trade-off between growth and reproduction during development. *Belgian Journal of Zoology* **134**: 41-45.

Nakadera, Y., J. M. Koene. 2013. Reproductive strategies in hermaphroditic gastropods: conceptual and empirical approaches. *Canadian Journal of Zoology* **91**: 367–381.

Norton, C. G. and J. M. Bronson. 2006. The relationship of body size and growth to egg production in the hermaphroditic freshwater snail, *Helisoma trivolvis*. *Journal of Molluscan Studies* **72**: 143–147.

Norton, C. G. and B. R. Newman. 2016. Growth, reproduction and longevity in the hermaphroditic freshwater snail *Helisoma trivolvis*. *Journal of Molluscan Studies* **82**: 178–186.

Norton, C. G., A. F. Johnson, R. L. Mueller. 2008. Relative size influences gender role in the freshwater hermaphroditic snail, *Helisoma trivolvis*. *Behavioral Ecology* **19**: 1122–1127.

Norton, C. G., A. F. Johnson, B. M. Nelson. 2018. The genetic basis of albinism in the hermaphrodite freshwater snail *Planorbella trivolvis*. *American Malacological Bulletin* 36: xx - xx.

Paraense, W. L. and L. R. Correa. 1988. Self–fertilization in the freshwater snails *Helisoma duryi* and *Helisoma trivolvis*. *Memorias do Instituto Oswaldo Cruz* **83**: 405–409.

Roff, D. A. 1992. *The evolution of life histories: theory and analysis*. Chapman and Hall, New York.

Schärer, L., T. Janicke, S. A. Ramm. 2014. Sexual conflict in hermaphrodites. *Cold Spring Harbor Perspectives in Biology* **7**: a017673.

Stearns, S. C. 1992. *The evolution of life histories.* Oxford University Press, Oxford.

Studier, E. H., K. E. Edwards, M. D. Thompson. 1975. Bioenergetics in two pulmonate snails, *Helisoma and Physa*. *Comparative Biochemistry and Physiology Part A: Physiology* **51**: 859-861.

Vianey-Liaud, M. 1989. Growth and fecundity in a black-pigmented and an albino strain of *Biomphalaria glabrata* (Gastropoda: Pulmonata). *Malacological Review* **22**: 25–32.

Vianey-Liaud, M., D. Joly, G. Dussart. 1996. Sperm competition in the simultaneous hermaphrodite freshwater snail *Biomphalaria glabrata* (Gastropoda: Pulmonata). *Journal of Molluscan Studies* **62**: 451–457.

Submitted: 14 June 2017; **accepted:** 30 January 2018; **final revisions received:** 2 February 2018