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The Relationship between Gall Size and Sizes of Selected Inhabitants of the Ball Gall of *Eurosta solidaginis* on *Solidago altissima*

Carol C. Mapes*, Jenna P. Ruoss, and Kelly Tessier

Biology Department, Kutztown University of Pennsylvania, Kutztown, PA 19530, USA
* Corresponding email: mapes@kutztown.edu

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ABSTRACT

The purpose of this study was to determine if there is a correlation between final ball gall mass and the mass of the gall inducing larva of *Eurosta solidaginis* and/or the mass of the adult fly that emerges, and to determine if there is a correlation between gall mass and the mass of the parasitoid *Eurytoma obtusiventris*. Another goal was to determine if there was a correlation between ball gall size and the sex of the adult fly that emerges. Adult flies were sexed and weighed as were the associated galls. Larval dry masses and dry masses of ball galls, with or without exit tunnels, were also determined. No correlation was found between ball gall mass and the mass of the adult fly that emerged from the gall. Adult female flies were of greater mass than male flies, but there was no correlation between gall mass and the sex of the gall former. Results indicate no biologically significant correlation between dry masses of the gall formers and dry masses of the galls. Furthermore, no biologically significant correlation was found between ball gall size and the size of the parasitic wasp *Eurytoma obtusiventris*. Interestingly, larger ball galls do not typically yield larger larvae or adults of *Eurosta solidaginis*.

Key words: Diptera, Tephritidae, *Eurosta solidaginis*, *Solidago altissima*, ball gall

Introduction

Ball gall formation (Figure 1A) on the tall goldenrod (*Solidago altissima* L., Asteraceae) is induced by the fly *Eurosta solidaginis* (Fitch) (Diptera: Tephritidae). Adults emerge from galls, mate and then the adult female fly deposits its fertilized egg into the terminal bud of *S. altissima* during late spring (Figure 1B). Once the larva hatches and tunnels into the stem, gall initiation begins. By the middle of September, both the larva and gall are considered full size. In autumn, larvae begin to scrape exit tunnels just up to the outer surface of the gall. The successful larva overwinters in the gall (Figure 1C), and then in spring, pupation occurs (Figure 1D), with the adult emerging in May in the northeastern United States (Uhler, 1951).

Larvae of *Eurosta solidaginis* may be jeopardized by the parasitoids *Eurytoma gigantea* Walsh (Hymenoptera: Eurytomidae) and *Eurytoma obtusiventris* Gahan

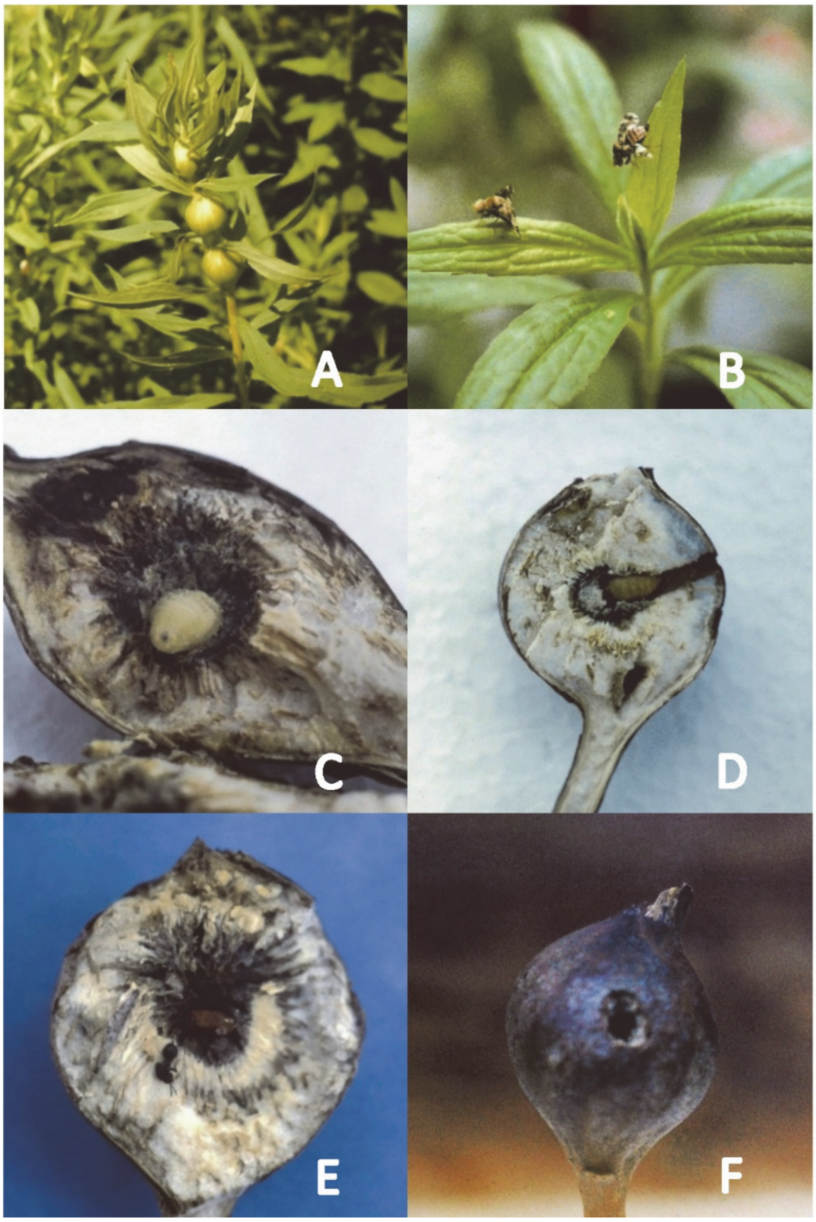

Fig. 1. A, goldenrod ball galls caused by *Eurosta solidaginis*; B, *Eurosta solidaginis* adults on *Solidago altissima*; C, third instar larva of *Eurosta solidaginis* inside ball gall; D, puparium of *Eurosta solidaginis* in ball gall with a completed exit tunnel; E, adult of *Eurytoma obtusiventris* that emerged from a premature puparium in a ball gall; F, goldenrod ball gall with a bird predation hole.

(Hymenoptera: Eurytomidae) (Figure 1E), by a beetle inquiline/predator *Mordellistena convicta* LeConte (Coleoptera: Mordellidae), bird predation (Abrahamson and Weis 1989) (Figure 1F), or predation by squirrels (Shealer *et al.* 1999). Typically, the parasitoid *Eurytoma gigantea* is found in smaller galls (Cane and Kurczewski 1976, Shealer *et al.* 1999). It has been shown that *E. gigantea* tends to be found in smaller galls because the adult wasps preferentially deposit eggs into smaller galls given the limited sizes of their ovipositors (Weis *et al*. 1985).

Downy woodpeckers, *Picoides pubescens* (Linneaus), and black-capped chickadees, *Poecile atricapillus* (Linneaus), are predators of *Eurosta solidaginis* during the winter months (Schlichter 1978). Bird predators have a tendency to seek out larger ball galls (Confer and Paicos 1985, Abrahamson and Weis 1989, Shealer *et al.* 1999). Abrahamson and Weis (1989) found that the combined effect of predatory birds preferentially seeking out larger ball galls and *Eurytoma gigantea* preferentially attacking smaller galls had a stabilizing effect on selection, causing *Eurosta solidaginis* in intermediate sized galls to have the highest survivorship. Similar findings have been reported for galls caused by *Diplolepis rosae* (Linneaus) on roses. Vertebrate predation frequency increased with gall size, while smaller galls had higher rates of attack by parasitoids (*László et al*. 2014).

Eurytoma obtusiventris is an internal parasite of *Eurosta solidaginis* (Uhler 1951) and it attacks *E. solidaginis* during the egg stage (Abrahamson and Weis 1997). *Eurytoma obtusiventris* causes the gall-former to make a premature puparium in late summer, and then it consumes its host and overwinters as a larva within the puparium prior to its pupation the next spring (Weis and Abrahamson 1985).

Given that birds preferentially seek out larger ball galls and that larger galls have the potential to provide more nutrition for the inhabitants, one might hypothesize that larger ball galls contain larger larvae of *Eurosta solidaginis*. The purpose of this study was to determine if there is a correlation between the sizes of ball galls and the sizes of fully developed *E. solidaginis* larvae within the galls as well as the sizes of *Eurytoma obtusiventris* inhabitants within ball galls. An additional goal was to determine if there is a correlation between the sizes of ball galls and the sizes of the *Eurosta solidaginis* adult flies that emerge from the galls. We aimed to answer the question as to whether larger ball galls contain larger *E. solidaginis* larvae, yield larger *E. solidaginis* adults, and contain larger *Eurytoma obtusiventris* inhabitants.

Materials and Methods

Goldenrod ball galls were collected from eleven *Solidago altissima* fields in Berks and Lehigh counties in Pennsylvania, USA over three different stages: late summer/early autumn (September 23~October 16, 2017), late autumn (November 11~December 7, 2017), and overwintering (February 24~April 28, 2017). Galls from the late summer/early autumn were collected prior to exit tunnel completion (pre-tunnel galls), and galls collected in late autumn had completed exit tunnels (galls with exit tunnels). The pre-tunnel galls and galls with exit tunnels were cut open, and inhabitants were identified. Galls and the respective inhabitants were dried in a Fisher Scientific Isotemp oven at 60°C for a minimum of 7 days, and dry masses of galls and inhabitants were determined using an ED244S Sartorius balance.

Eurosta solidaginis adults were reared from overwintering galls that were placed in mesh covered mason jars or *Drosophila* culture tubes fitted with foam inserts and incubated in a Conviron E7/2 growth chamber at 22°C under a 14-hour photoperiod. *E. solidaginis* flies were sexed, and fresh masses were determined using an ED244S Sartorius balance within 24 hours of emergence. Adult flies and their respective galls were dried in a Fisher Scientific Isotemp oven for 7 days, and dry masses were also recorded.

Once the dry masses for galls and inhabitants had been recorded, these masses were plotted using Microsoft Excel, and linear regressions were performed to determine if there was a correlation between gall sizes and sizes of gall inhabitants. Regression analyses were also used to determine if results were statistically significant. Means and standard errors were calculated to make comparisons of the sizes of *E. solidaginis* larvae, adults, and ball galls. Two-sample independent t-tests were conducted to determine if the mean dry masses of *E. solidaginis* larvae for pre-tunnel galls and exit tunnel galls were significantly different. Two-sample t-tests were also used to determine if the mean sizes of female and male adult *E. solidaginis* flies were significantly different and to determine if their associated galls were significantly different in mass. F-tests were run prior to all t-tests to determine whether or not there was equality of variances.

Results

There was no significant relationship between the dry masses of fully formed pre-tunnel ball galls and the dry masses of their respective *Eurosta solidaginis* larvae (R^2= 0.0319, $F_{1,41}$=1.35, p=0.25); larger galls did not yield larger larvae for the late summer/early autumn time period (Figure 2). There was a very

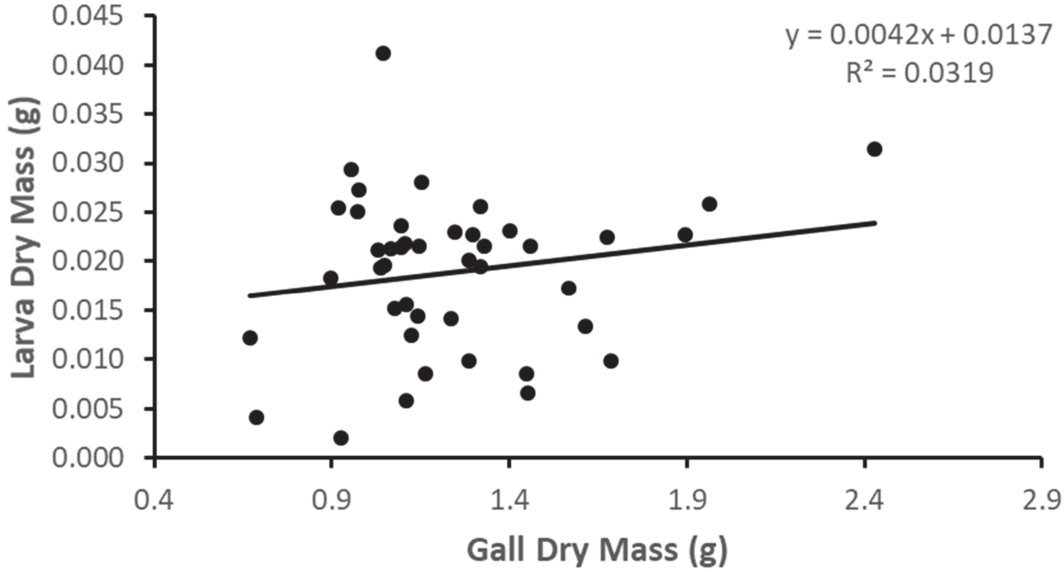

Fig. 2. The gall dry mass and respective larval *Eurosta solidaginis* dry mass for the late summer/early autumn time period showed no significant relationship (R^2=0.0319, $F_{1,41}$=1.35, *p*=0.25). These larvae had not started their exit tunnels.

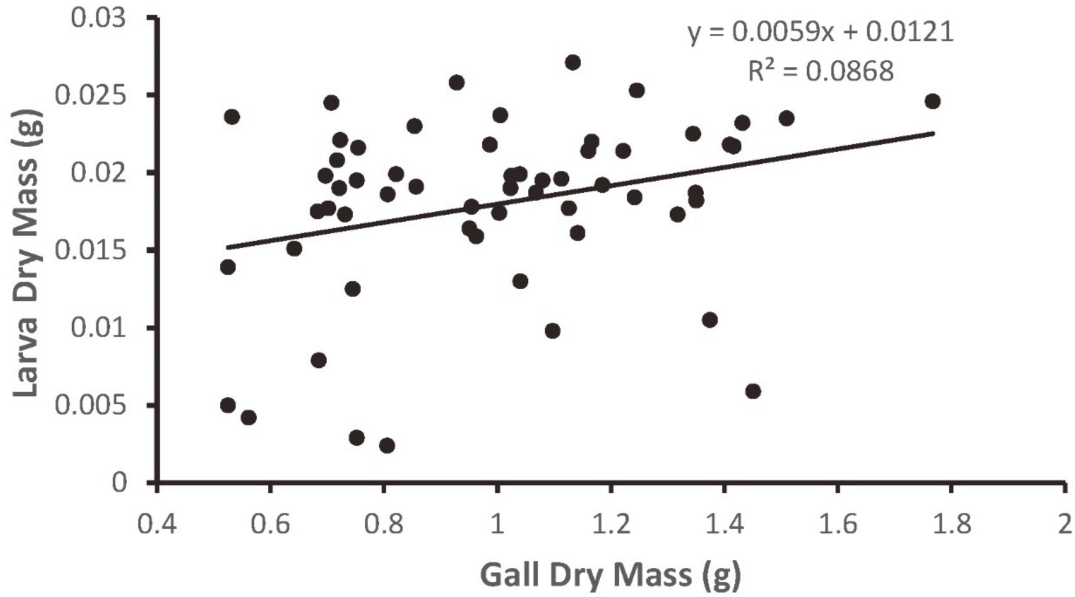

Fig. 3. The gall dry mass and respective larval *Eurosta solidaginis* dry mass for the late autumn collection showed a very weak positive correlation (R^2=0.0868, $F_{1,56}$=5.32, *p*<0.05). At this collection stage, larvae had completed exit tunnels.

weak, but statistically significant, positive relationship between the dry masses of galls with completed exit tunnels and the dry masses of the associated *E. solidaginis* larvae for the late autumn time period (R^2=0.0868, $F_{1,56}$=5.32, *p*<0.05; Figure 3). The mean dry masses of the *E. solidaginis* larvae from fully formed pre-tunnel galls was 18.9 ± 1.2 mg (n=43) while the mean dry masses of the larvae from the galls with exit tunnels was 18.0 ± 0.8 mg (n=58), showing no significant difference (t=0.67, df=73, *p*=0.25).

No relationship was found between the dry masses of the adult *E. solidaginis* flies and the dry masses of their respective galls (R^2=0.0154,

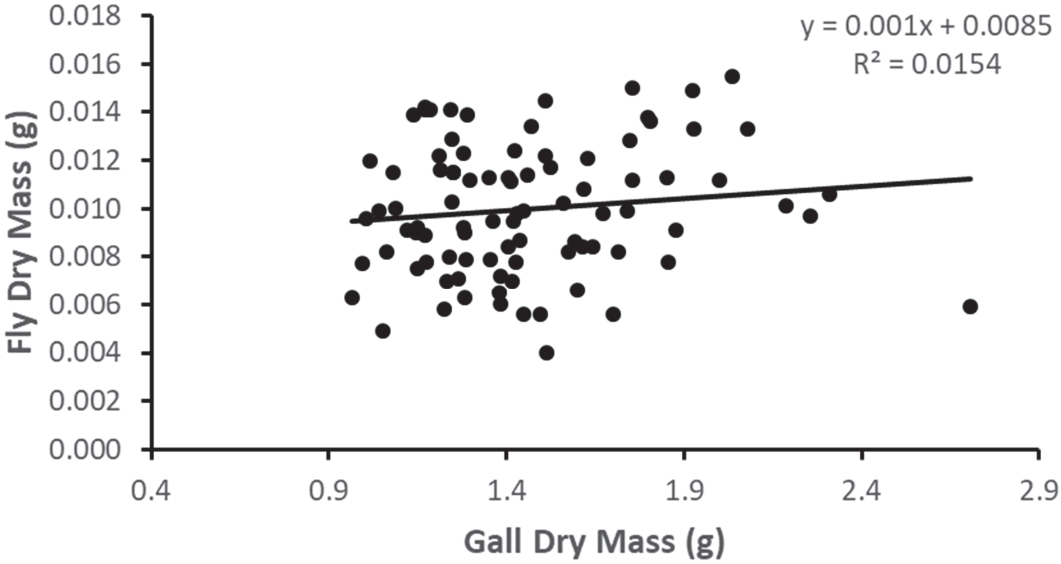

Fig. 4. The gall dry mass and the adult *Eurosta solidaginis* fly dry mass exhibited no significant relationship (R^2=0.0154, $F_{1,88}$=1.38, *p*=0.24).

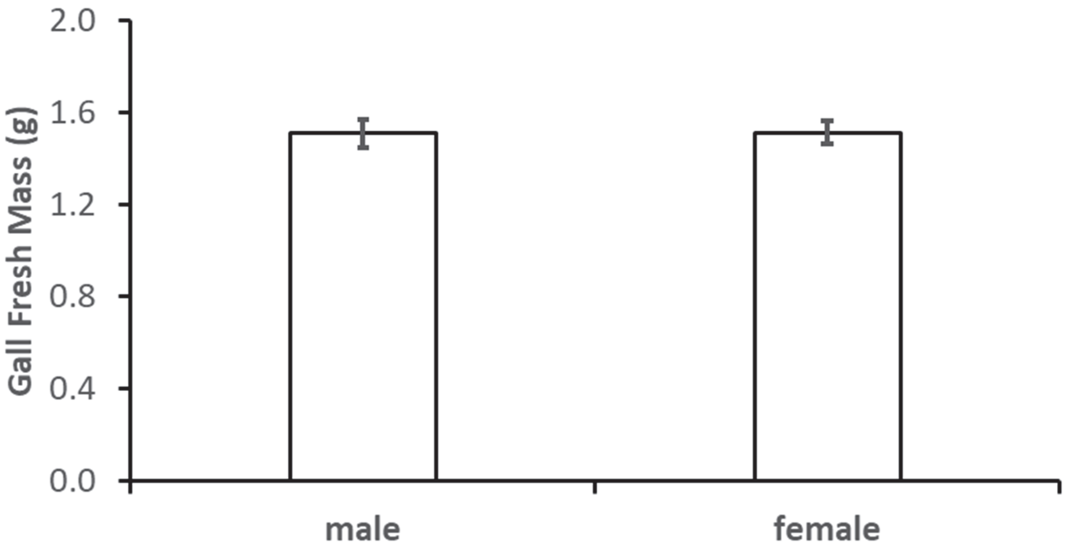

Fig. 5. The mean fresh masses of galls caused by male *Eurosta solidaginis* flies (n=51) and female *E. solidaginis* flies (n=43). Standard error bars and a two-sample independent t-test indicated that results were not significantly different (t=-0.06, df=92, *p*=0.48).

$F_{1,88}$=1.38, *p*=0.24; Figure 4). The mean fresh mass of female *E. solidaginis* flies was significantly greater at 29.8 ± 0.8 mg (n=54) compared to males flies with a mean of 24.7 ± 0.7 mg (n=57) (t=4.77, df=109, *p*<0.01). However male galls and female galls were not statistically different in mass (t=-0.06, df=92, *p*=0.48; Figure 5).

The late autumn collection of galls parasitized by *Eurytoma obtusiventris* showed a weak, but statistically significant, positive relationship between the dry masses of the galls and the dry masses of *E. obtusiventris* (R^2= 0.1792, $F_{1,27}$=5.89, *p*<0.05; Figure 6).

Discussion

For pre-tunnel galls, there was no relationship between ball gall dry mass and *Eurosta solidaginis* larval dry mass, while for galls with exit tunnels, there was only a very weak positive relationship between ball gall dry

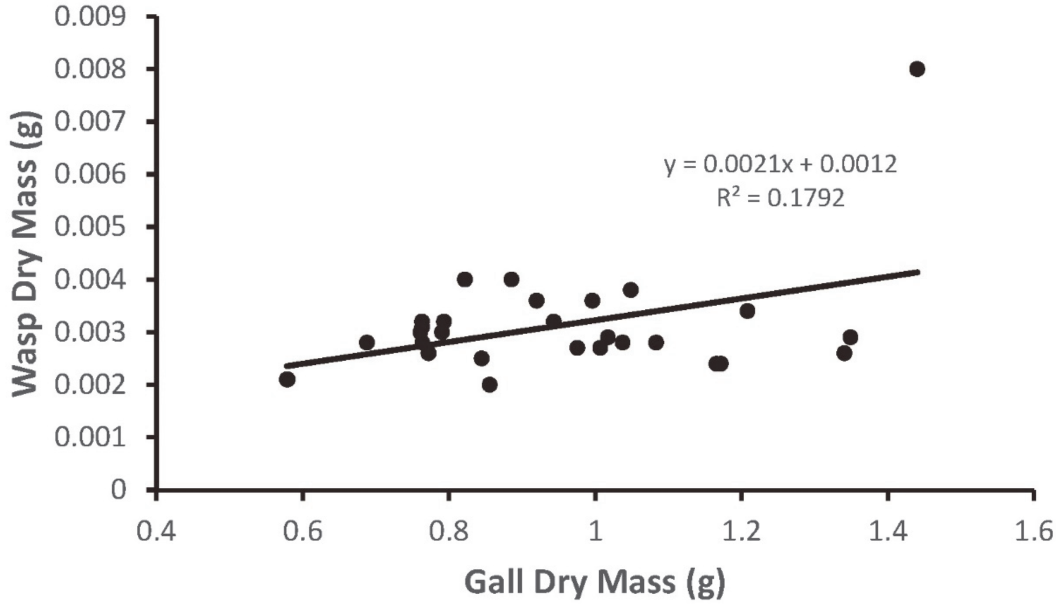

Fig. 6. The gall dry mass and the dry mass of the parasitoid *Eurytoma obtusiventris* showed a slightly positive correlation for the galls collected in late autumn (R^2=0.1792, $F_{1,27}$=5.89, p<0.05).

mass and larval dry mass with an R^2 of 0.0868. This suggests that gall size is not a reliable predictor of the size of the *E. solidaginis* larval inhabitant. Fully formed ball galls were collected at the two different stages, before and after exit tunnel completion. If gall size were an important factor affecting *E. solidaginis* larval size, one might have predicted that ball galls with exit tunnels would be more likely to show a strong correlation between gall size and larval size. If the larvae gain nutrition during the process of scraping exit tunnels through the gall tissue, larger ball galls would have more tissue that would need to be removed/consumed in the process of exit tunnel completion resulting in larger larvae. Instead the results of this study did not show a strong correlation between ball gall size and *E. solidaginis* larval size for galls with completed tunnels. Interestingly mean larval mass did not increase or change from the pre-exit tunnel collection to the completed exit tunnel collection time suggesting that *E. solidaginis* larvae do not gain mass during exit tunnel completion. Any nutrition that is gained may be used to provide energy for the exit tunnel completion process as there was no loss of mass in the larvae as well.

How *et al.* (1993) reported a significant positive correlation between *E. solidaginis* larval dry mass and ball gall dry mass with an R^2 of 0.284 when doing a combined analysis of fully developed ball galls found on *S. altissima* and *S. gigantea* from Minnesota and Pennsylvania. It is not clear whether these galls had completed exit tunnels at the time of collection but the regression presumably included a mix of what is now known as two host races of *E. solidaginis* (Abrahamson and Weis 1997). It is interesting that there was a stronger positive correlation between gall size and *E. solidaginis* larval size than what was obtained in this study in which we report no correlation for pre-tunnel galls and only a weak positive correlation with an R^2 of 0.0868 for galls with completed exit tunnels. Perhaps *S. gigantea* host race galls show more of a correlation between *E. solidaginis* larval masses and ball gall masses. Cane and Kurczewski (1976) indicated they found no significant correlation between the diameters of ball galls and the *E. solidaginis* larvae within, but did not provide data to support their statements.

The results presented here show a weak positive correlation between the dry masses of *Eurytoma obtusiventris* and the dry masses of their associated ball galls (Figure 6). Given that *E. obtusiventris* is an internal parasite of *Eurosta solidaginis* during the entire development of the ball gall, it is not surprising that the results are similar to those that we report for *E. solidaginis* in showing, at most, a weak positive correlation between ball gall mass and inhabitant mass. Cane and Kurczewski (1976) indicated they found no significant correlation between the diameters of ball galls and *Eurytoma obtusiventris* larvae within, but did not provide any supporting data.

There was no correlation between the dry masses of the galls and the dry masses of the adult *E. solidaginis* flies that emerged from the galls in this study. Contrary to what one might have expected, larger ball galls did not yield larger *E. solidaginis* adults. The findings reported here are in contrast to those of Stille (1984) who found a correlation between the sizes of adults of the gall former *Diplolepis rosae* and the sizes of the rose galls from which they emerged.

Female *Eurosta solidaginis* adults were shown to be of greater mass than male *E. solidaginis* adults in this study in corroboration with findings of Hess *et al*. (1996). Despite their larger size, the females did not emerge from larger galls. This is unlike some gall systems where females produce larger, morphologically distinct galls (Goncalves *et al*. 2005).

Results presented here indicate that larger ball galls did not yield larger adults of *Eurosta solidaginis.* In addition, gall size is not a good predictor of the sizes of *E. solidaginis* larvae or the sizes of the parasitoid *Eurytoma obtusiventris*. While one might have assumed that larger galls produce larger *Eurosta solidaginis* larvae and yield larger adults, this was not clearly shown in this study. These results could be explained by considering that larvae of *E. solidaginis* do not consume all of the surrounding gall tissue, and that the wasps of *Eurytoma obtusiventris* are internal parasitoids of the gall former and do not consume any gall tissue. As a result, larger galls do not act as a source of a larger amounts of food for these inhabitants.

Birds such as woodpeckers seek out larger *Eurosta solidaginis* galls (Abrahamson and Weis 1997), but the results of this study indicate that the largest ball galls will not always yield the largest larvae of *E. solidaginis*. Shealer *et al.* (1999) reported that when all occupants were considered, including the parasitoids and gall formers, the masses of ball gall occupants in smaller galls were significantly less than the masses of ball gall occupants in medium and larger ball galls, and small galls were more likely to be empty. Given that *Eurytoma gigantea* more frequently parasitizes smaller ball galls and that *E. gigantea* larvae weigh much less than *Eurosta solidaginis* larvae, bird predators would be more likely to find a larger inhabitant to consume in larger ball galls (Shealer *et al.* 1999). Shealer *et al*. (1999) also found that the eastern gray squirrels (*Sciurus carolinensis* Gmelin) that predated on ball galls did not prefer ball galls of a particular size class, and they were found to consume *E. solidaginis* larvae as well as parasitoids.

While birds are more likely to find an inhabitant in a larger ball gall, and are more likely to find an *E. solidaginis* larva when avoiding smaller galls, the results of this study show that birds that seek out larger ball galls are missing the large *E. solidaginis* larvae that can be found in smaller galls. The results presented here, along with results from other studies, could also have practical implications for ice fishermen. Ball galls are sometimes collected by individuals to remove the *E. solidaginis* larvae to use as bait for ice fishing (Frank Mapes, personal communication). If the goal is to find large *E. solidaginis* larvae, one should not restrict oneself to collecting large galls given that there is not a strong positive correlation between ball gall size and the sizes of *E. solidaginis* larvae.

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北美一枝黃花上癭蠅（***Eurosta solidaginis***）的球狀蟲癭大小和癭內生物之相關性

Carol C. Mapes*, **Jenna P. Ruoss, and Kelly Tessier**

Biology Department, Kutztown University of Pennsylvania, Kutztown, PA 19530, USA
* 通訊作者 email: mapes@kutztown.edu

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摘　　要

本研究之目的為瞭解成熟的球狀蟲癭重量是否與癭蠅(*Eurosta solidaginis*)之幼蟲重量及/或與成蟲重量有相關性，及蟲癭重量是否與癭蠅寄生蜂 *Eurytoma obtusiventris* 重量相關，另一目的則為探討蟲癭大小是否與癭蠅成蟲之性別有相關性。首先鑑定成蟲性別，再進行成蟲及蟲癭的稱重，同時記錄幼蟲乾重、蟲癭乾重與是否有羽化通道。結果顯示蟲癭重量和成蟲重量之間無相關性，雌蟲較雄蟲重，但蟲癭重量和性別無相關性。由此顯示癭蠅之造癭者的乾重和蟲癭的乾重無生物學之顯著關聯，再者，蟲癭大小和寄生蜂體型也無生物學中的顯著相關，其中有趣的是，較大的蟲癭並不會孕育出較大的癭蠅幼蟲或成蟲。

關鍵詞：雙翅目、果實蠅科、*Eurosta solidaginis*、*Solidago altissima*、球狀蟲癭