

Location and seasonal differences in adult dragonfly size and mass in northern Mississippi, USA (Odonata: Libellulidae)

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ABSTRACT

Size and mass are often uniformly related within individuals and populations, but the relationship may vary in time or space. I asked whether isolated adult dragonfly populations within the same environmental context (climate, physiography, ecoregion) differ in both size and mature mass, and whether earlier emerging dragonflies are both larger and heavier on average. Differences were apparent among locally separated populations (ca 130-160 km apart), with the most northerly populations containing larger and heavier adults on average. Site-level environmental variation probably exerted a larger influence than broad-based ecogeographic rules (e.g., Bergmann's rule) at this fine scale. On average, earlier emerging dragonflies tended to outsize and/or outweigh later emerging dragonflies, a commonly observed pattern in adult odonates and other insect taxa. Size and mass did not produce the same results in every case, suggesting the size-mass relationship within dragonfly species can vary among spatially or seasonally isolated adult populations.

INTRODUCTION

Uniform patterns of mass at length are well documented from composite samples of terrestrial and aquatic insects (Rogers et al. 1976; Smock 1980; Sample et al. 1993; Benke et al. 1999; Johnston & Cunjak 1999; Sabo et al. 2002). In most insects, size attained during the larval growth period is directly related to adult size (Rowe & Ludwig 1991). Adult dragonfly skeletal size is fixed, but individual mass varies with sclerotization, dehydration, foraging behavior, and other factors over the lifespan (Corbet 1999: 264-271, 370-375). Nevertheless, mature adult dragonfly mass may scale linearly with size when measured across individuals belonging to the same population (Sabo et al. 2002; Bried et al. 2005). The relationship may vary, however, among dragonfly populations separated in

time or space (Bried & Ervin 2007). Therefore, it is not clear whether mass will follow size patterns between spatially disjointed populations or will decline from early to late emerging broods; insect size at emergence typically declines as the reproductive season progresses (Vannote & Sweeney 1980; Mousseau & Roff 1989; Falck & Johansson 2000).

Given that size and mature mass are often related within dragonfly populations and that adult size is fixed while mass is not, I revisited a previously published data set (Bried & Ervin 2007) with the following questions: (1) Do isolated populations within the same environmental setting (climate, physiography, ecoregion) differ in both size and mature mass on average? (2) Are earlier emerging dragonflies both larger and heavier on average? This study takes Bried & Ervin (2007) a step further by analyzing size and mass separately instead of together in a regression framework. In the previous paper there were two possible scenarios underlying significant main effects or intersecting regression slopes: (1) size and mass both varied statistically, but to different degrees, across the location or seasonal gradient, or (2) one measure varied while the other did not; the analysis in Bried & Ervin (2007) did not decide which one. Direct analysis of size and mass in this paper clarifies whether size, mass, or both varied in each case. Also note that not every case in Bried & Ervin (2007) met the assumptions of parametric analyses and therefore results in these cases were potentially spurious. Another new piece of information provided here is the direction of the time effect, i.e., whether June or August contained the larger dragonflies on average.

MATERIAL AND METHODS

Dragonflies (Anisoptera) were collected from three locations in the northern half of Mississippi, USA: North Mississippi Refuges Complex (NMRC; approximately centered at 33.78°N, 90.15°W), Noxubee National Wildlife Refuge (NNWR; 33.43°N, 88.82°W), and Strawberry Plains Audubon Center (SPAC; 34.80°N, 89.42°W). All three study locations lay within the North Central Hills physiographic region and the Upper East Gulf Coastal Plain ecoregion. The NMRC also bordered the Mississippi River Alluvial Plain ecoregion and the Loess Hills and Delta physiographic regions, lying 129.9 km from NNWR and 131.3 km from SPAC. Separation between NNWR and SPAC (156.9 km) represented the entire latitudinal gradient (1.4° difference) for the study. Across the state monthly mean temperatures ranged from ca 2.0 to 33.5°C, and high temperatures may exceed 32°C for up to 100 d per year. Most of the state received 125-175 cm mean annual rainfall (Krotzer et al. 2008).

Five species were targeted for collection: *Erythemis simplicicollis* (Say), *Libellula incesta* Hagen, *Libellula lydia* Drury, *Pachydiplax longipennis* (Burmeister), and *Perithemis tenera* (Say). As adults these are among the most commonly encountered odonate species in Mississippi (Krotzer et al. 2008). Collecting took place during part of the early summer dragonfly flight season (3-11 June) and part of the late summer flight season (16-23 August) in 2004. Each species was collected from the same site in a given study location during both collection periods. Habitat included an oxbow depression, farm ponds, moist-soil managed marsh, beaver marsh, and reservoir littoral zone. All specimens of a species were collected from a single wetland rather than from multiple wetland sites of a study location. Processing and analysis were limited to males with hard integuments, reproductive coloration, and fully transparent wings (Anholt et al. 1991). Right hind wing length (nearest 0.1 mm) was measured from point of attachment to the distal margin using digital calipers.

Table 1. Cell frequencies (number of dragonflies) available for randomization tests. Cells with “< 20” were excluded from analysis. NMRC: North Mississippi Refuges Complex; NNWR: Noxubee National Wildlife Refuge; SPAC: Strawberry Plains Audubon Center.

Species	NMRC		NNWR		SPAC	
	June	August	June	August	June	August
<i>Erythemis simplicicollis</i>	34	33	28	27	33	32
<i>Libellula incesta</i>	< 20	< 20	28	29	32	23
<i>Libellula lydia</i>	< 20	21	< 20	< 20	22	27
<i>Pachydiplax longipennis</i>	43	40	40	43	43	40
<i>Perithemis tenera</i>	33	20	< 20	< 20	< 20	< 20

Specimens were dried for 24 h at 70°C, stored in a dessicator, and weighed to 0.01 mg on a Fisher Scientific accu-124D electronic balance. See Bried & Ervin (2007) for more details on data collection and processing methods.

Adult dragonflies are strong dispersers but I assumed that ca 130-160 km spacing of locations exceeded their flight distances (none of these species are migratory), and despite being relatively long-lived I assumed the 65 d gap between collection periods exceeded their typical longevity (Corbet 1980: 199). Therefore, I assumed that scales in this study ensured that adult populations were independent yet still within the same environmental setting and annual flight period.

Three species (*L. incesta*, *L. lydia*, *P. tenera*) were more difficult to find or catch than initially anticipated, resulting in gaps in the test gradients (Table 1). Random data permutation methods (Edgington & Onghena 2007) were used to compare average dry mass and wing length among factor levels (2 time periods, 3 locations). For single-factor cases (June vs August in *L. lydia* and *P. tenera*; NMRC vs SPAC in *L. lydia*), observations were shuffled throughout the matrix of specimens and available factor levels. The difference between means in the original data was used as a nondirectional test statistic, or effect size. For two-factor comparisons, meaning the 2 x 2 (*L. incesta*) and 2 x 3 (*E. simplicicollis*, *P. longipennis*) arrays from Table 1, main effects were analyzed by shuffling observations within levels of one factor and between levels of the other factor (Edgington & Onghena 2007). This parsimonious approach required just four tests in each species, including one for the location effect (randomize within time periods and among locations) and one for the time effect (randomize within locations and among time periods) repeated for weight and size. A nondirectional test statistic was given by summing the observations across levels of one factor, squaring each sum, dividing each squared sum by the number of observations, and then totaling across the levels (Edgington & Onghena 2007). Both test statistics were re-computed over randomizations, and the number of re-computed statistics equal to or exceeding the test statistic was taken as the significance level. I used Resampling Stats (written by S. Blank, ©2005; Resampling Stats Inc., Arlington, Virginia) and 10,000 iterations for all significance testing.

Randomization tests were used instead of parametric statistics because of unequal sample sizes and inconstant residual error variance found in this data set (Bried & Ervin 2007). Another reason is that the ‘p-value’ in a randomization test measures the strength of evidence for an effect (Manly 1997), and should be less prone to sample size and arbitrary hypothesis statements (see Johnson 1999; McBride 2001).

RESULTS

Regarding the first question, isolated populations in the same environmental setting differed in size and/or mass in most cases (Table 2). The most northerly populations (SPAC) contained the largest and heaviest dragonflies on average (Table 2), ignoring mass in *Pachydiplax longipennis*. Regarding the second question, earlier emerging dragonflies tended to outsize and/or outweigh later emerging dragonflies on average (Fig. 1). Aggregate size losses ranged from 0.5% (*Libellula incesta*) to 7.1% (*Perithemis tenera*) and weight losses from 3.5% (*P. longipennis*) to 16.4% (*L. lydia*) between the June and August collection periods. In the seasonally segregated adult populations different statistical conclusions (using $\alpha = 0.05$) for size and mass were reached in three species (Fig. 1), and in the populations segregated by location differences were seen in two species (Table 2).

DISCUSSION

Regarding the first question, site-level environmental variation (e.g., food availability) probably held a larger influence than broad-based ecogeographic rules (e.g., Bergmann's rule), given that the populations were separated within a small spatial extent (ca 130-160 km) and shared environmental setting. The larger and heavier northern populations may have been driven by coincidence and not Bergmann's rule, as the study locations were arranged nearly in an equilateral triangle and not a north-south alignment, and because gradual and persistent change in size (i.e., clines) may not apply at such a small spatial scale. It seems more reasonable that insects follow converse Bergmann's rule and adjust the number of generations per growing season (Roff 1980). Interestingly, the couple studies of broad-scale size clines in adult odonates found U-shaped patterns (Johansson 2003; De Block et al. 2008).

Regarding the second question, changes in temperature, photoperiod, and possibly other cues (e.g., food supply, predation risk) may have signaled increased reproductive costs as the flight season progressed, advancing the onset of emergence by accelerating physiologic development at the expense of additional somatic growth (Johansson & Rowe 1999). This phenomenon is supported in adult odonate populations collected in the wild (Michiels & Dhondt 1989; Falck & Johansson 2000) or reared experimentally (Plaiستow & Siva-Jothy 1999; Johansson et al. 2001). Other studies have reported size decreases with advancing season in the same species as analyzed here (Cothran & Thorp 1982; Koenig & Albano 1987).

This study was descriptive and did not control for factors that affect odonate size and mass patterns. Exact age of the specimens was unknown, although I tried to standardize based on qualitative observations of body hardness, wing transparency, and color (Anholt et al. 1991). Body weight of adult odonates changes over the flight season (Koenig & Albano 1987; Uéda 1989; Dunham 1993) but also over shorter periods depending for instance on food availability and weather conditions (Grabow & Ruppell 1995; Corbet 1999: 370-371). As such, collecting at > 2 time points over the flight season may have been a better albeit more labor-intensive strategy. I further acknowledge that samples at given sites may have included mixed native and immigrant individuals. Despite these uncontrolled factors and sources of natural variability, differences were still detected beyond what was expected

Figure 1: Average size and mass of adult dragonfly populations in the early summer (3-11 June) versus late summer (16-23 August) flight season in northern Mississippi, USA. The effect was analyzed by shuffling observations within the locations and between the time periods. Cases with fewer than 20 specimens (see Table 1) were excluded. Strength of evidence is shown where $p < 0.05$. Note that *Erythemis simplicicollis* and *Libellula lydia* overlap considerably in the upper graph.

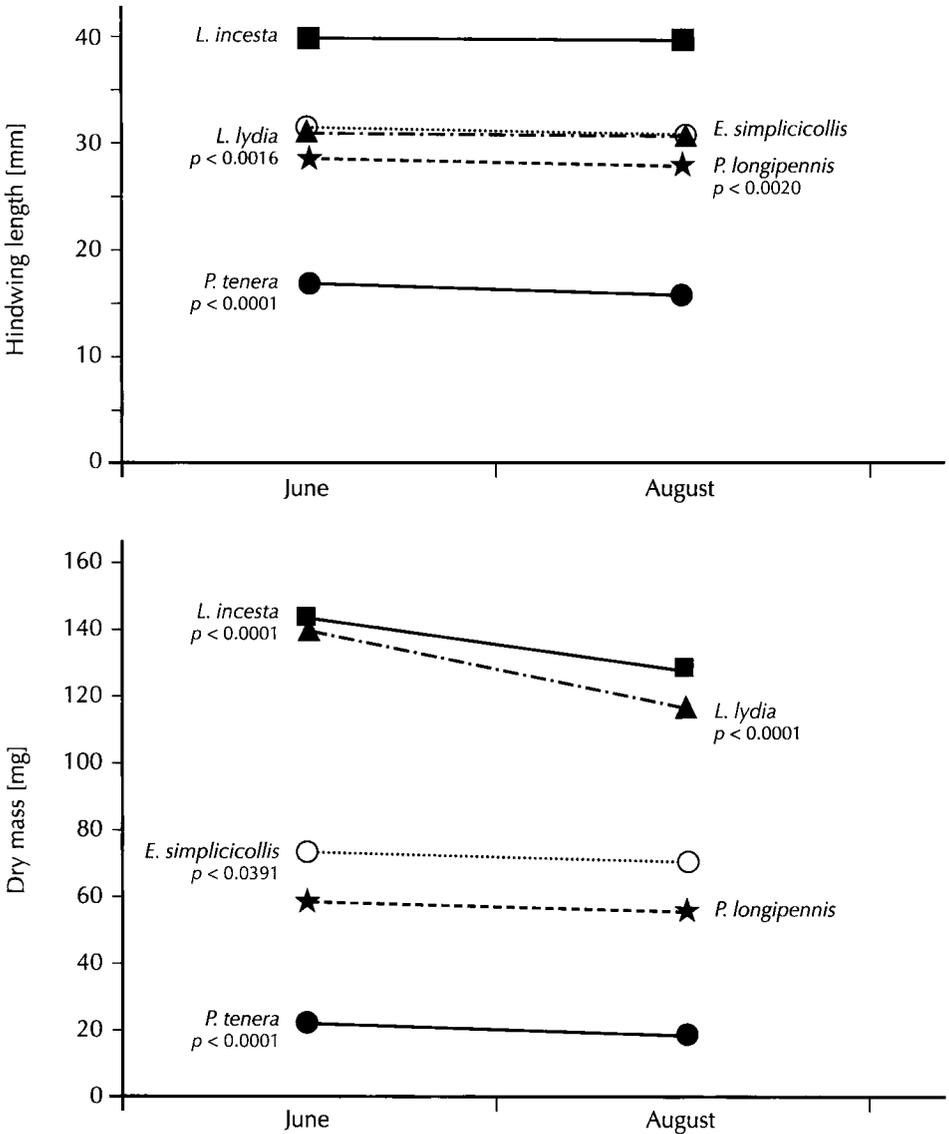


Table 2. Average size (hind wing length in mm) and mass (dry weight in mg) \pm standard deviation of adult dragonflies at three locations in northern Mississippi, USA. Strength of evidence (p) derives from randomization tests with 10,000 iterations. The effect was analyzed by shuffling observations within the collection periods (June, August) and between the locations. n.a.: not analyzed (< 20 specimens; see Table 1). NMRC: North Mississippi Refuges Complex; NNWR: Noxubee National Wildlife Refuge; SPAC: Strawberry Plains Audubon Center.

Species		NMRC	NNWR	SPAC	p
<i>Erythemis simplicicollis</i>	Size	30.9 \pm 1.3	30.8 \pm 1.2	31.2 \pm 1.1	0.2176
	Mass	69.0 \pm 13.7	72.6 \pm 12.7	75.6 \pm 9.2	0.0069
<i>Libellula incesta</i>	Size	n.a.	39.6 \pm 1.3	40.1 \pm 1.2	0.0365
	Mass	n.a.	130.4 \pm 17.9	140.5 \pm 15.5	0.0016
<i>Libellula lydia</i>	Size	30.8 \pm 0.8	n.a.	31.4 \pm 0.7	0.0063
	Mass	116.9 \pm 9.7	n.a.	139.5 \pm 9.3	< 0.0001
<i>Pachydiplax longipennis</i>	Size	27.9 \pm 1.5	28.2 \pm 1.9	28.5 \pm 1.5	0.0259
	Mass	54.5 \pm 12.3	59.2 \pm 6.6	58.4 \pm 12.1	0.0640

by chance alone. Of course statistical significance is not the same as biological significance and testing scientific hypotheses (Johnson 1999, 2002). For example, it is not clear whether statistically significant differences of a few milligrams or tenths of millimeters, as seen in this study, matter to dragonfly fitness (cf. Sokolovska et al. 2000).

Different conclusions for size vs mass (based on randomization tests) may have contributed to heterogeneity in size-mass regression slopes found in previous analyses over the same spatiotemporal gradient (Bried & Ervin 2007). In those analyses, significant interaction between location (or time) and size suggested that mass varied with size differently between locations (or times). However, this could have resulted from significant but variable change in mass and size together, or, significant change in one measure but not the other. For example, slopes differed between June and August in *Libellula incesta* and *Perithemis tenera* (Bried & Ervin 2007: table 4), but according to the current analysis *L. incesta* declined only in mass whereas *P. tenera* declined in both mass and size (Fig. 1).

As a volumetric quantity it stands to reason that individual mass of aquatic macroinvertebrates can increase roughly as a cubic function of length during growth (Benke et al. 1999). Mass may also scale uniformly with length when measured across individual adult dragonflies within populations (Sabo et al. 2002; Bried et al. 2005). However, this linear size-mass relationship within dragonfly species can vary among spatially or seasonally isolated adult populations.

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