Biodiversity, Phenology, and Thermoregulatory Strategies of Odonates at Pierce Cedar Creek Institute

Undergraduate Research Grants for the Environment Report

Elaine M. Sheikh and Dr. Matthew Douglas

Grand Rapids Community College

October, 2012

ABSTRACT: Forty-three species of dragonflies from five families and sixteen species of damselflies from three families were identified at Pierce Cedar Creek Institute in Hastings, Michigan (latitude 42.6459 and longitude -85.2908) between May 7 and August 10, 2012. Our study showed that Pierce Cedar Creek Institute provides habitat to a greater number and variety of odonates than expected. The diurnal phenology of the odonates varied by species, with smaller and medium dragonflies generally out earlier in the day and active into the afternoon, and large dragonflies are more active near dusk. We found that dragonflies and damselflies use a variety of active and passive thermoregulatory strategies. We found that the mean Δ T (the difference between ambient and thoracic temperature) as well as the heating /cooling curves and preferred flight temperatures, are positively correlated with increasing thoracic size of the odonate. In addition, we found that the flow of haemolymph from the wings to the thorax does not function to significantly regulate thoracic temperature.

Key Words: Odonata, anisoptera, zygoptera, thermoregulation, facultative endothermy, biodiversity, phenology

INTRODUCTION

Dragonflies and damselflies are commonly thought to be ectothermic organisms; however, they have some capability for endothermy (May 1974), and thus are more accurately classified as facultative endotherms. These are organisms whose internal body (thoracic) temperatures are partly dependent on thermal conditions, but can also employ endothermy through muscular thermogenesis over a broad range of thermal environmental conditions.

When ambient thermal conditions are lower than the threshold required for controlled flight, odonates lack the energy necessary for flight and may be unable to fly in a coordinated manner (May 1974). This means that valuable time for feeding and mating is lost while the partially debilitated insects (vulnerable to predation) wait for ambient thermal conditions to rise to a sufficient level necessary to support controlled flight.

Dragonflies and damselflies employ a variety of behavioral and physiological methods to regulate thoracic temperature (Heinrich 1981, 1993, May 1974, 1995); which is significant because the thorax is where the flight and locomotor muscles reside, therefore, is the body segment that must maintain minimum muscle temperature necessary to support flight activity. These strategies can be behavioral and physiological such as wing-whirring and frantic feeding (May 1995). Wing-whirring is a process of synchronously contracting the thoracic flight muscles to vibrate the wings and thus producing heat through muscular thermogenesis in a process similar to shivering, a strategy most common in "flyers;" odonates that spend the majority of their time in active flight (Heinrich 1993). In contrast, "frantic feeding," is gliding interspersed with sporadic flight as if hunting, but without predatory intentions (May 1995). Biophysical strategies of thermoregulation may also be utilized, such as choosing strategic resting/basking postures (dorsal, lateral, or oblique, for example) to maximize or minimize exposure and subsequent absorption of radiation from the sun and immediate surrounding environment (May 1974). In addition, members of five families of odonates have been shown to change body coloration by either increasing melanization, thereby increasing the absorption of radiation from the sun, or decreasing melanization to increase reflection of solar radiation to prevent overheating (Heinrich 1993).

Behavioral thermoregulatory strategies may also be employed to prevent overheating when ambient thermal conditions rise to extreme levels. These cooling strategies include tactics such as reducing flight intensity and wing beat frequency to avoid expending excess energy from wing movement during flight, thereby reducing heat production from rapidly contracting thoracic muscles (May 1974), "water-dipping" (Silsby 2001), color changes (Conrad and Pritchard 1989), and strategic body postures such as obeslisking, a basking position where the abdomen is held nearly perpendicular to the thorax to reduce exposure to and absorption of radiation from the sun (May 1974).

Three main variables directly affect the thoracic temperature of an odonate: thoracic size/mass, climate, and strategies of thermoregulation (May 1974, Willmer 1983). For facultative endotherms, the thermal environment or "thermal climate space" is composed of four main variables: air temperature, wind speed, insolation, and humidity. Thermoregulatory strategies adapt to changes in these variables and define virtually all life activities for active flying insects such as odonates, as well as lepidopterans (Casey 1980, Douglas 1978), hymenopterans (Stone 1993), and coleopterans (Casey 1981). Because of their preferred thermoregulatory strategies and subsequent behavior in the field, some species are collectively considered perchers – individuals which spend the majority of their time basking – while others are referred to as flyers – individuals which spend the majority of their time in flight (Heinrich 1993, May 1974). Thermoregulatory strategies also determine the time of day at which odonates can be active. Species that heat faster are able to take to the air sooner than their slower-heating competitors, and are able to begin feeding, mating, and escaping predators earlier in the day, securing more territory, prey, and breeding locations for themselves (Douglas 1978). Most species are only able to fly during the day; however, some larger species are able to hunt at night (Kirschbaum 2007).

Body temperature is an important factor that affects metabolic rates and most physiological processes mediated by enzymes. For this reason, an organism's thermoregulatory mechanisms will directly affect those metabolic activities that determine its survival and reproductive success. For the purpose of studying the effects of the radiant environment on odonates' temperature, organisms are treated as black bodies (Gates 2003). The larger the surface area of an organism is, the more radiation will be absorbed by that organism (however, the emittance area of an organism increases with increasing surface area as well). A smaller odonate will absorb less energy from solar radiation than a larger odonate, and will also have more surface area relative to volume from which to lose heat via convection, so smaller insects will lose proportionally more heat through convection than larger specimens (May 1974).

Thoracic size/mass also plays an integral part in the thermoregulatory techniques utilized by a particular odonate species (May 1974). Species with greater mass should have a greater flight metabolism and produce more heat endothermically by muscular processes, demonstrating more effective thermoregulation than smaller species. The muscle mass of damselflies, for example, may be too small to produce any measureable increase in body temperature, whereas a larger libellulid or aeshnid dragonfly may produce a great amount of heat by internal metabolic processes. Because of this, larger insects should have a higher maximum flight temperature than a smaller insect; in fact, large specimens might quickly overheat and be forced to abandon flight early in the day if the thermal conditions are extreme.

Like all organisms, dragonflies and damselflies are not only coupled to their thermal microenvironment, they must also utilize basic physical mechanisms for survival and reproduction. The modes of energy exchange between a dragonfly and its thermal environment are theoretically straightforward, but are complicated under field conditions because of variations in body morphology, anatomical structures, and physiological and behavioral thermoregulatory strategies. For example, the *characteristic dimensions* (e.g., "diameter") of an insect may be increased by the presence of wings that reduce convective heat loss by modifying the flow of air over the insect's body while maximizing the absorption of solar radiation close to the thorax.

A complete analysis of the behavioral and biophysical thermoregulatory strategies of odonates would require a detailed knowledge of the physical interactions between dragonflies and damselflies and their immediate thermal environment under field conditions. This study is directed toward elucidating these thermoregulatory strategies as much as possible.

In addition to creating a synoptic collection, establishing an analysis of the diurnal activity and seasonal phenology of species and measuring diversity, this study attempted to compare and contrast the variations and relative efficiency of the thermoregulatory strategies exhibited by anisopterans and zygopterans representative of Pierce Cedar Creek Institute. Some research has been done concerning thermoregulation and behavior of some of the larger dragonfly species, such as *Anax junius*,the common Green Darner, (May 1974) and several of the larger species of libellulids. However, little research has been conducted concerning the thermoregulatory strategies of damselflies. The only comprehensive works published are those concerning the temperature at which physiological color morphs take place (Conrad and Pritchard, 1989). In addition, we were unable to find any research conducted on smaller species of dragonflies. We investigated several of these under-studied groups to provide a more complete comparison of thermoregulatory techniques, behavior, and environmental niches in different group of odonates. We hypothesized that a) there would be a large amount of odonate species diversity, b) diurnal phenology would vary significantly between differently sized odonate species, c) larger odonates would utilize more active thermoregulatory strategies than smaller odonates, which would utilize primarily passive strategies of thermoregulation, d) Δ T will be larger in large odonates than in smaller individuals, and e) haemolymph flow from wing to thorax will have no significant effect on thoracic thermoregulation.

METHODS

I. Biodiversity and Phenology Study: Sampling took place from May 7 to August 10, 2012. Phenology analysis was performed by daily walking each trail at PCCI at different times of day (between 9 am and 9 pm) and by performing a qualitative analysis of species diversity, relative abundance, and distribution in each separate habitat/location on the Institute. Select species were netted using a butterfly net, placed in insect envelopes or collection jars, and taken back to the laboratory for identification. Several identification guides as well as Internet sources were used to identify specimens. Specimens were killed using 70% ethyl alcohol, then pinned, dried, and labeled to create a synoptic collection that will be left at PCCI for educational purposes.

Most captured specimens were keyed out and released; a few were kept for a synoptic collection, for laboratory experimentation, or for the study of heating and cooling curves of dead specimens. In addition, we compared our findings to the Checklist of Michigan Odonata (created by the University of Michigan and Mark O'Brian of the Michigan Odonata Survey, 2004.) Locations of sightings and/or capture were estimated using a GPS system providing coordinates accurate to within 3.5 meters of capture.

II. Measurement of General Thermal Climate: Data from the weather stations at PCCI were used to measure wind speed, air temperature, insolation, and humidity at the same time as insect thoracic temperatures are measured. We also used a portable pyrometer to record the immediate thermal environment of the insect to create a thermal climate space for different size categories of odonates in a later paper).

III. Measurement of Thoracic Temperature: Thoracic temperatures of odonates in flight or at rest were measured in insects under field and laboratory conditions. Insects in the field were netted and quickly probed in the mesothorax with a fine gauge copper-constantan thermocouple that provided an instantaneous measurement of thoracic temperature accurate to 0.1 ˚C using the standard "grab-and-stab" methodology (May 1974).

IV. Measurement of Heating and Cooling: Live, freshly killed, and dried specimens were heated and cooled in the laboratory under controlled ambient temperatures and radiation provided by a 100-Watt full-spectrum incandescent bulb at a height that delivered 2 calories/m^2 of energy to the specimen. This amount of radiation approximated the insolation of sunlight at noon on a clear July day. The bulb was suspended at the same height for all specimens. Specimens were allowed to cool naturally unless otherwise noted. Convective heat loss was kept to a minimum by performing experiments in the absence of wind or drafts. The odonates' heating and cooling curves under controlled thermal conditions were recorded every five seconds.

Size Categories:

Insects were measured using a small caliper and measuring tape marked in millimeters. Small dragonflies were classified as specimens with approximately 0.35 cm diameter of mesothorax, 0.80 cm-long thorax, and a 1.3 cm-long abdomen. Medium dragonflies were classified as those with approximately 0.65 diameter of mesothorax, 1.0 cm-long thorax, and a 2.0 cm-long abdomen. Large dragonflies had approximately a 0.7 diameter of mesothorax, 2.0 cm-long thorax and 4.8 cm-long abdomen.

V. Statistical Analysis of Δ T: Statistical analysis of Δ T (ambient temperature – thoracic temperature) for size-matched species was used to determine if thermal niche portioning took place. The null hypothesis was that species of the same mass will have similar average Δ T values. We hypothesized that species with different thoracic diameter may actually split up the thermal environment differently, and therefore have statistically different average Δ T values (statistical analysis was determined using the Kruskal-Wallis test and the assistance of Dr. Sango Otieno of Grand Valley State University's Statistics Counseling Center).

VI. Determination of Variables Involved in Thermoregulation:

Select captured specimens were placed into one of three groups: group A had their wings removed completely; group B had their wings ablated from the thorax but then repressed in a normal position so that they physically touched the insect's thorax (this prevented haemolymph flow from wings to thorax while maintaining the characteristic dimensions and structural integrity of the insect. There is a possibility that haemolymph could leak from the insect's thorax through the wing stumps, however, upon post-experimental examination, this was not noticed); group C was the control group: insects with wings left intact. Thoracic temperature was measured as a heating and cooling curve over a 120 second period of time. This determined the extent to which wing venation and haemolymph flow from the wings to the thorax affected thoracic temperature.

Specimens were separated into five different groupings based on thoracic size: large dragonflies, medium dragonflies, small dragonflies, large damselflies, and small damselflies (see measurement classifications on page 4). The thermoregulatory strategies were observed and thoracic temperatures of each group were measured in the field and rates of heating and cooling were tested in the laboratory under controlled conditions to determine the extent to which the size of the thorax affected temperature regulation.

Specimens were observed as to whether or not physiological color changes take place; temperatures of color change were to be measured and data compiled to determine what factors affected temperature-related color morphs.

RESULTS

Biodiversity Study:

We found 43 species of Suborder Anisoptera from 5 different families: 8 species from Aeshnidae, 5 species from Corduliidae, 2 species from Cordulegastridae, 6 species from Gomphidae, and 21 species from Libellulidae. We also found 16 species of Suborder Zygoptera from 3 different families: 2 species from Calopterygidae, 4 species from Lestidae, and 10 species from Coenagrionidae. See appendix for a complete species list.

Phenology Study:

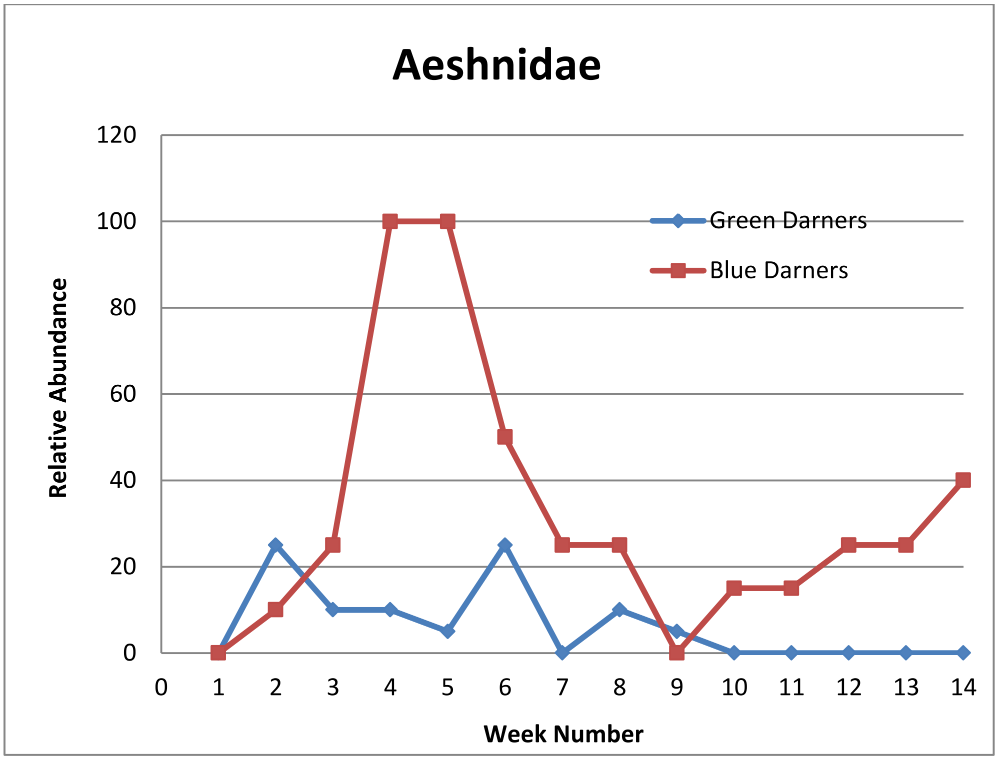

Figure 1. Relative abundance of Aeshnidae dragonflies observed at PCCI between May 7 (Week 1) and August 6 (Week 14) in 2012.

Beginning around May 10th, migrating aeshnids were sighted flying over the prairies near the Brewster Lake, Cedar Creek, Tall Grass Prairie, and Old Farm Trails. They were most active between 12 p.m. and 4 p.m., flying about 3.5 meters above the ground. Populations peaked in mid- to late May. Around May 24th, as the average ambient temperature and day length increased, the aeshnids were inactive throughout the day and would emerge from 6 p.m. to 9 p.m. During this time they would fly in the prairies in swarms of approximately 60 to 100 darners about 2 meters above the ground. This flight pattern continued until the end of June, when ambient temperatures spiked to 38 ºC, causing a sharp decline in population numbers and flight activity. A stray darner or two would occasionally be found flying or basking in the edges of woodlands adjacent to the prairies. The blue darner population slowly began picking up in mid- to late July, not reaching the status of moderately common sightings until early August when temperatures dropped back to more reasonable highs of 27-29 ˚C and migratory species began their migration south from their northern summer breeding grounds.

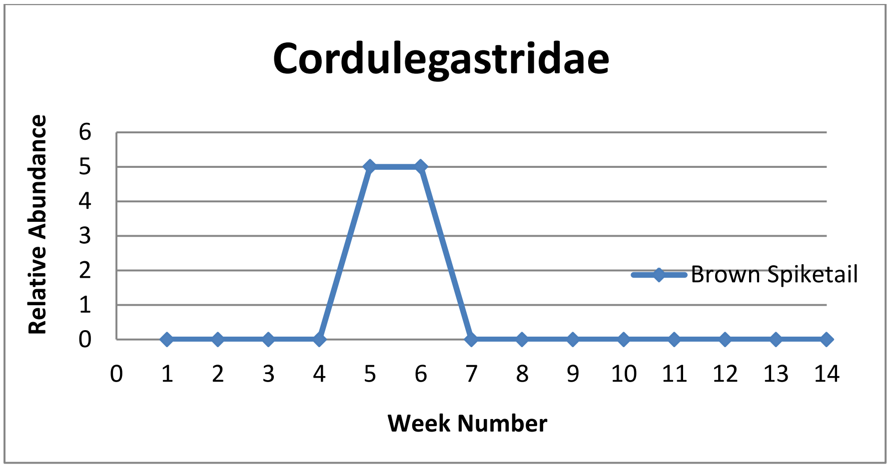

Figure 2. Relative abundance of Cordulegastridae dragonflies observed at PCCI between May 7 (Week 1) and August 6 (Week 14) in 2012.

Only one species of corduligastrid – the Brown Spiketail – was identified at PCCI. It was only observed in isolated cases in clearings in the woods on the Brewster Lake Trail the first and second full weeks of June.

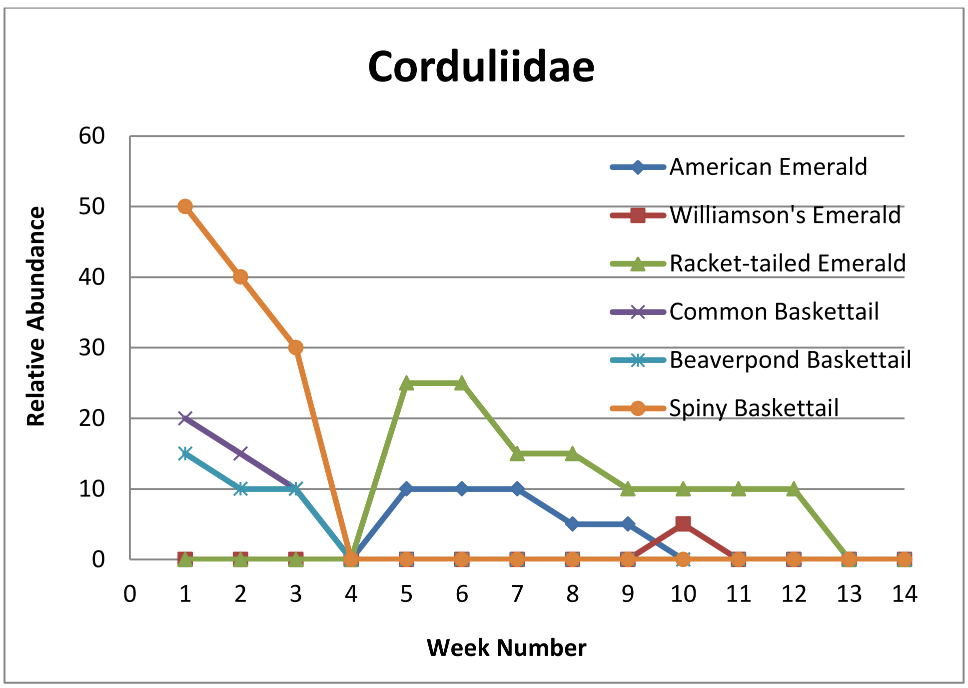

Figure 3. Relative abundance of Corduliidae dragonflies observed at PCCI between May 7 (Week 1) and August 6 (Week 14) in 2012.

In early May, there was a large number of corduliids at PCCI, comprising three species of baskettails (Beaverpond, Common, and Spiny Baskettails). These species inhabited woodlines on the prairies and small clearings in the woods. The Spiny Baskettail was the dominant species on the Institute the first two weeks of the study (May 7th – May 20th). These specimens were most active between 10 a.m. and 4 p.m.

The overall corduliid populations decreased to obscurity in mid-June, when the baskettails were replaced with American, Racket-Tailed and Williamson's Emeralds, which populated the woodlines, prairies and woods at PCCI until the first week in August.

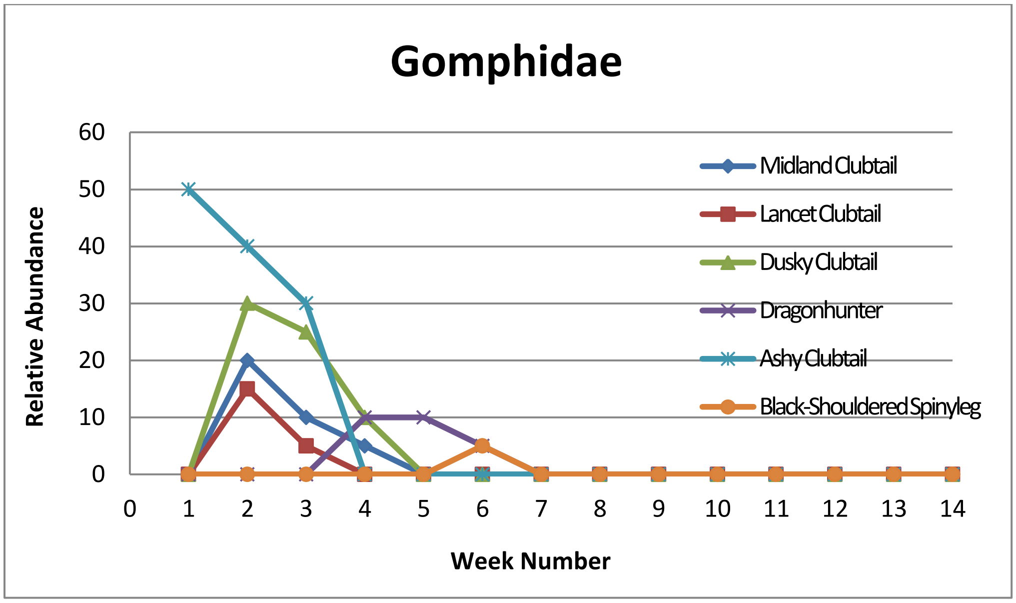

Figure 4, Relative abundance of Gomphidae dragonflies observed at PCCI between May 7 (Week 1) and August 6 (Week 14) in 2012.

The first gomphids were observed the second week of May. These were all Clubtails. The Dragonhunters appeared at the very end of May but their populations declined in only three weeks. Black-shouldered Spinylegs were only sighted at PCCI for one week. The gomphids were most active from 12 p.m. to 4 p.m. Population numbers dropped drastically correlating with the heat wave starting in late June.

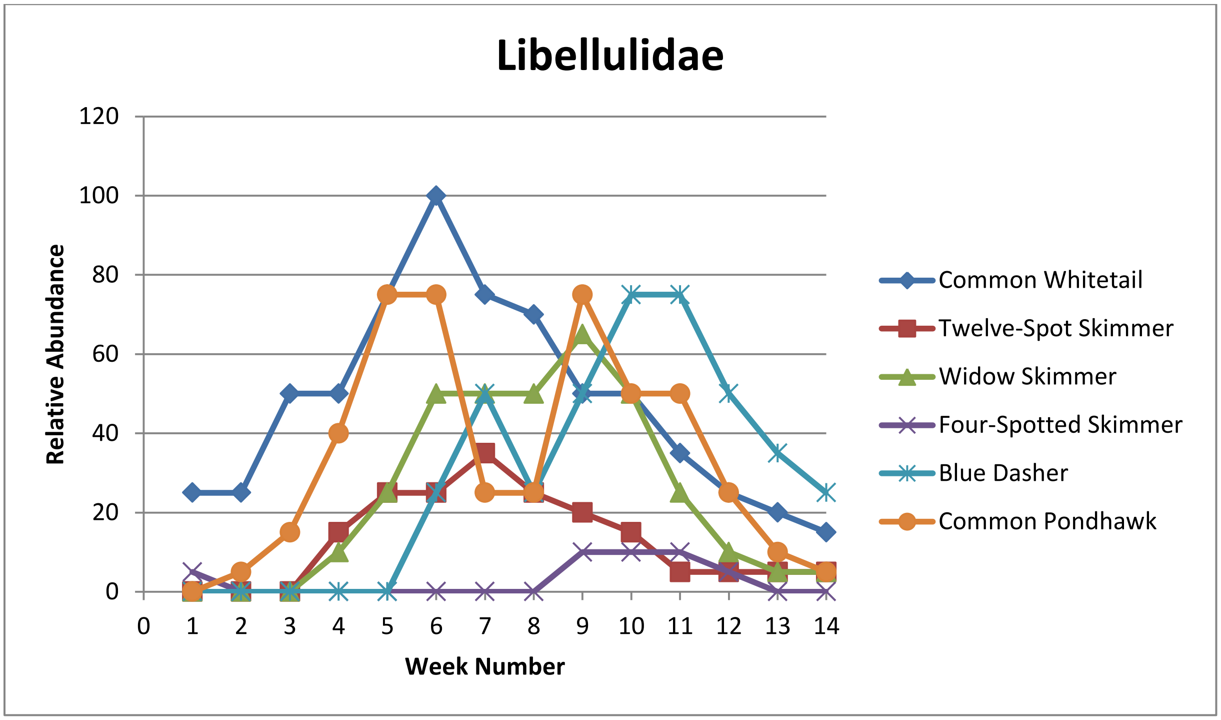

Figure 5a: Relative abundance of Libellulidae dragonflies observed between May 7 (Week 1) and August 6 (Week 14) in 2012.

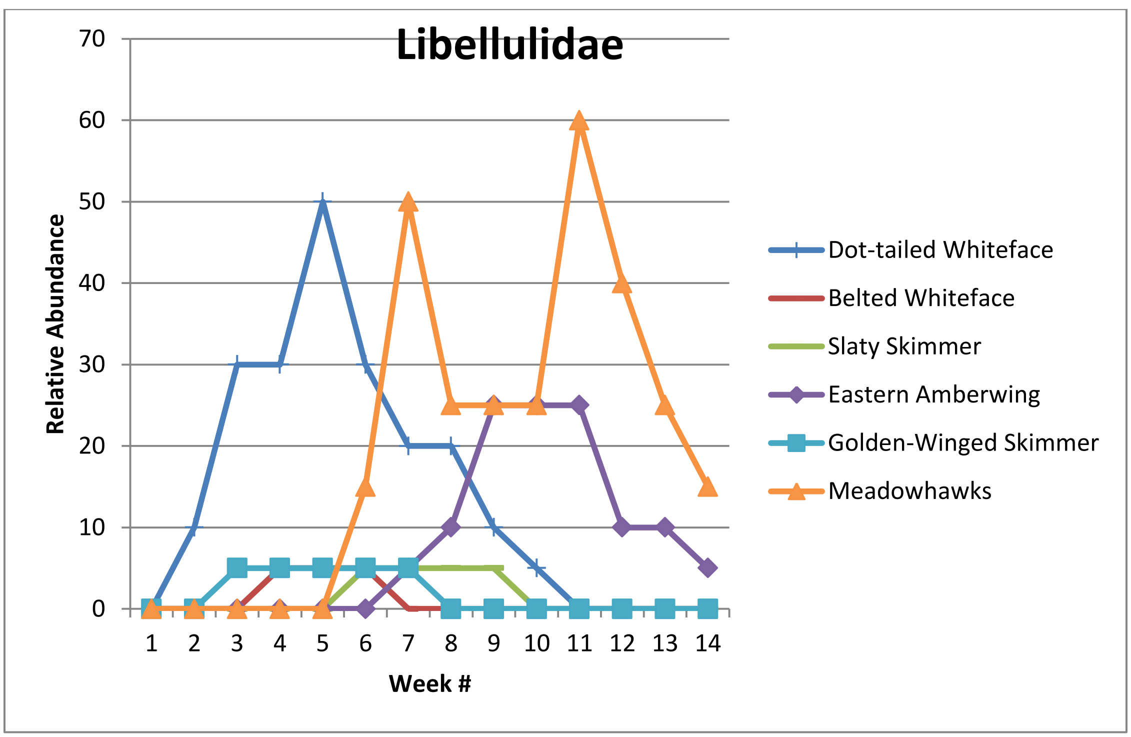

Figure 5b: Relative abundance of Libellulidae dragonflies observed between May 7 (Week 1) and August 6 (Week 14) in 2012 continued.

The Libellulidae had the greatest biodiversity and abundance of any dragonfly family at PCCI and the dominant species of odonate each week was member of the genus libellulidae. The general characteristics of each species' phenology were as follows:

King Skimmers (Libellula): The skimmers were the first libellulids to be sighted at PCCI and maintained active populations throughout the summer. These odonates were active during most of the day, usually from 10 a.m. until 4 p.m., but stayed active as late as 8 p.m. on especially hot days, when overheating could be a problem. The Common Whitetail, found in all habitats on the institute (prairies, woodlines and clearings in the woods, and wetlands) became active the second week of May. Their peak abundance was the second week of June. Their population gradually decreased but remained active at PCCI through August. In addition, the Common Whitetail was the dominant species weeks 4-7 of the study. The Twelve-Spot and Widow Skimmer appeared at PCCI the last week in May, an odonate-rich period of time. They inhabited the prairies and wetland areas of the Institute, and their populations lasted the duration of the summer. The Slaty Skimmers appeared on PCCI's prairies and wetlands the first week of June, and their population held steady until mid-July, when it waned slightly and held at a lower density through August. The last species, the Four-Spotted Skimmers, had the same habitat as the other king skimmers. This species had a single specimen identified the second week of May. Then none were sighted until the first week of July. They remained on the Institute throughout July and disappeared in the beginning of August.

Meadowhawks (Sympetrum): Seven species of Meadowhawks were identified on the prairies at PCCI. Due to the fact that newly emerged and female specimens can only be definitively identified in the laboratory the Meadowhawks in our study were all grouped together as Sympetrum. The first Sympetrum were identified the third week of June. Populations peaked during the last week of June, decreased as the heat wave intensified in early July, and then peaked again the last week of July, experiencing a steady decrease but sizable population through August. These libellulids were active primarily from 11 a.m. to 3 or 4 p.m.

Amberwings (Perithemis): The Eastern Amberwing was the only species of Perithemis on the prairies of PCCI. Individuals were first noted around June 20th. The population peaked between the first and third week of July, and decreased by the end of July but remained present on the Institute through August. The Amberwings were usually sighted obelisking in the prairies between 12 p.m. and 3 p.m.

Blue Dasher (Pachydiplax): These species were first sighted in prairies, woodlines, and clearings in woodlands the second week of June, populations of this species peaked the third and fourth weeks of July, with population numbers remaining fairly high through the rest of the summer. It was the dominant species Weeks 12-14 of the study. Blue Dashers were active between 10 a.m. and 4 p.m.

Pondhawks (Erythemis): The Common Pondhawk was one of the PCCI's dominant species of odonate. It appeared the third week of May and the population was still strong through August. Peak population was during the second and third weeks of June. This species inhabited all of PCCI's habitats and was the dominant species Week 3 and Weeks 8-11 of the study. These odonates were also active between 10 a.m. and 4 p.m.

Rainpool Gliders (Pantala): Only the Wandering Glider was identified on PCCI's prairies. Isolated sightings occurred in late July/early August, and took place in mid-afternoon (not enough sightings were made to approximate the full range of peak activity times).

Whitefaces (Leucorrhinia): Two whiteface species were identified: the Dot-tailed Whiteface and the Belted Whiteface. The Dot-tailed Whiteface was the more prevalent of the two species. The first Dot-tailed Whiteface specimen was identified the third week of May, and the population swelled rapidly to peak during the second and third weeks of June. The population diminished and was gone the second week of July. Belted Whiteface individuals were only sighted a few times, all in the first three weeks of June. The Whitefaces inhabited a large variety of prairie, woodland, and wetland habitats and were primarily active between 10 a.m. and 4 p.m.

Small Pennants (Celithemis): One species from the genus Celithemis, the Halloween Pennant, was sighted on the PCCI's prairies. Individuals were sighted starting in the last week of June; the population peaked the second week of July, and disappeared the first week of August. The Halloween Pennant was most active between 11 a.m. and 3 p.m.

Thermoregulatory Study

Average Heating and Cooling Curves (in the laboratory):

Specimens that were representative of their respective size categories and showed an average heating and cooling curve for their size categories were used to create the figures in this section.

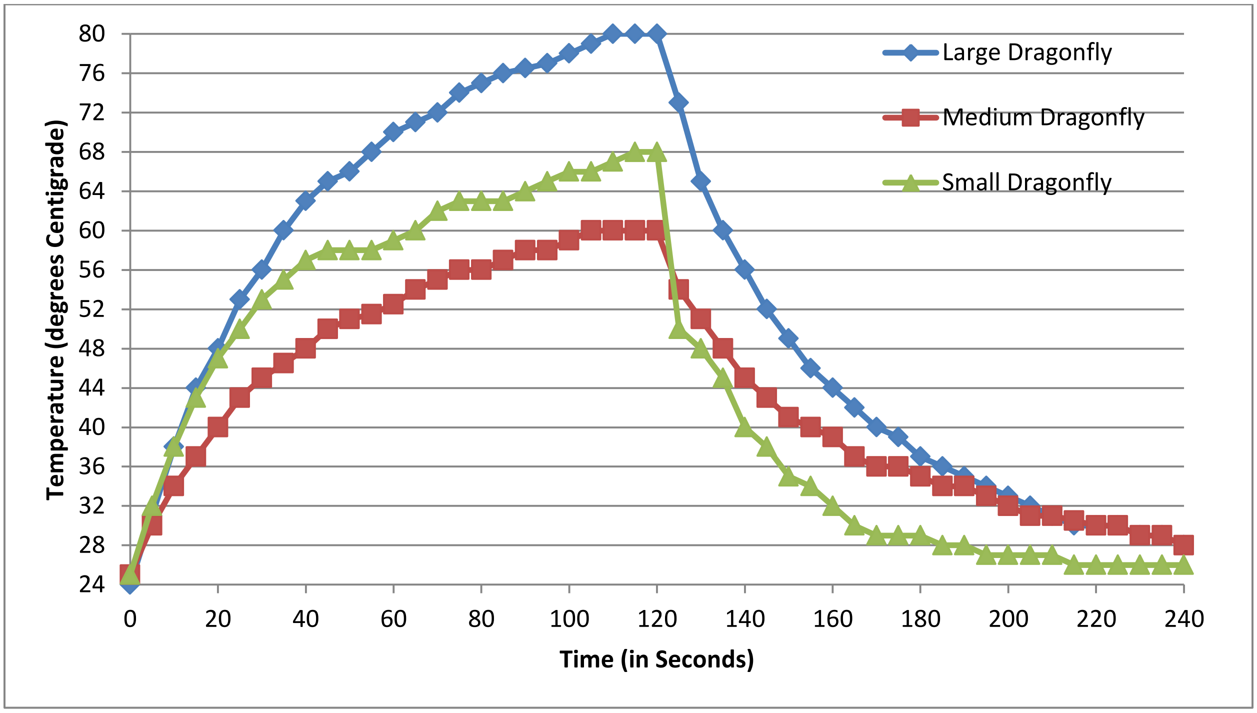

Figure 6 – comparison of the heating and cooling curves of large, medium and small dragonflies in the dorsal basking position.

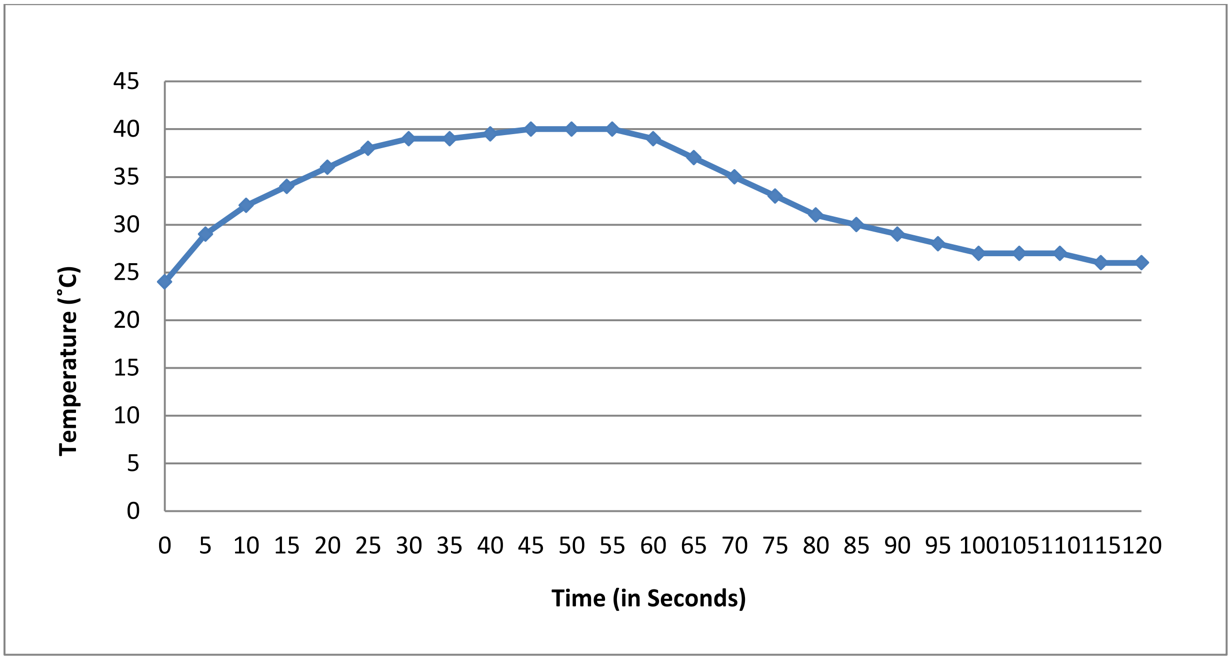

Figure 7 – heating/cooling curve of a dead Dot-tailed Whiteface (small dragonfly) arranged in the obelisk basking position

The dead Dot-tailed Whiteface (small dragonfly), *Leucorrhinia intacta*, shows a decreased slope of the heating and cooling curve when in the obelisk position compared to the dorsal bask position (see figure 6).

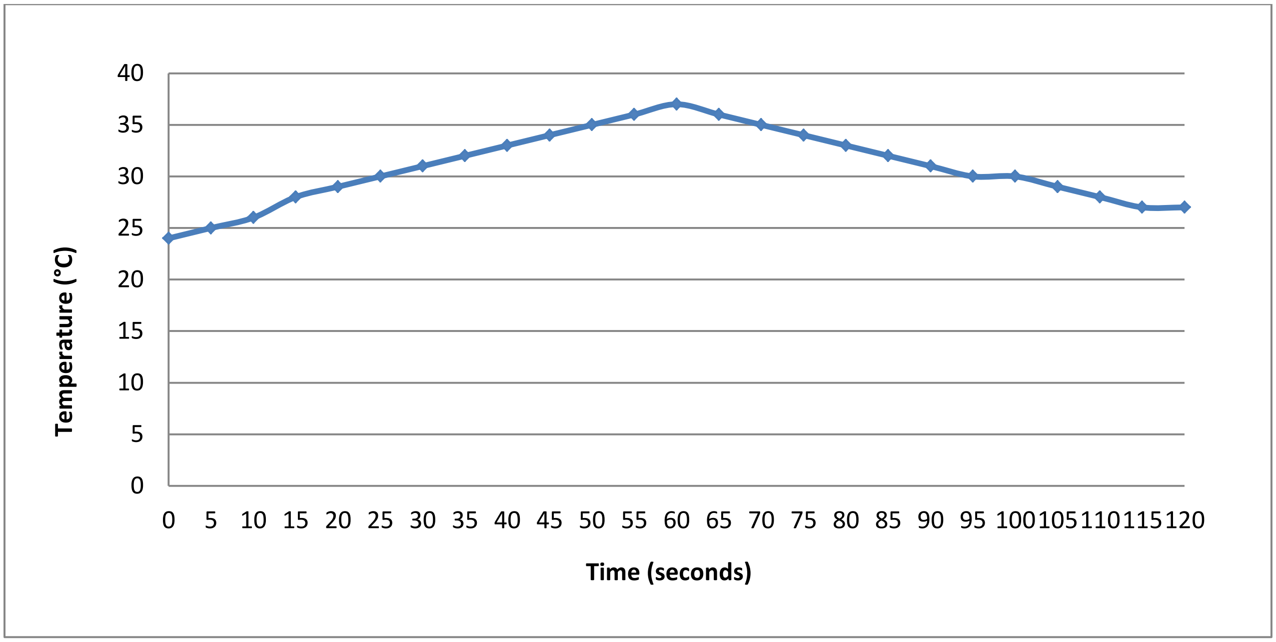

Figure 8 – heating/cooling curve of a dead Green Darner (large dragonfly) arranged in a vertically basking position.

The dead Green Darner (large dragonfly), *Anax Junius*, arranged in a vertically hanging basking position showed a much slower heating curve and reached a lower maximum temperature than did the dorsally basking Green Darner (Figure 6).

Large Damselfly:

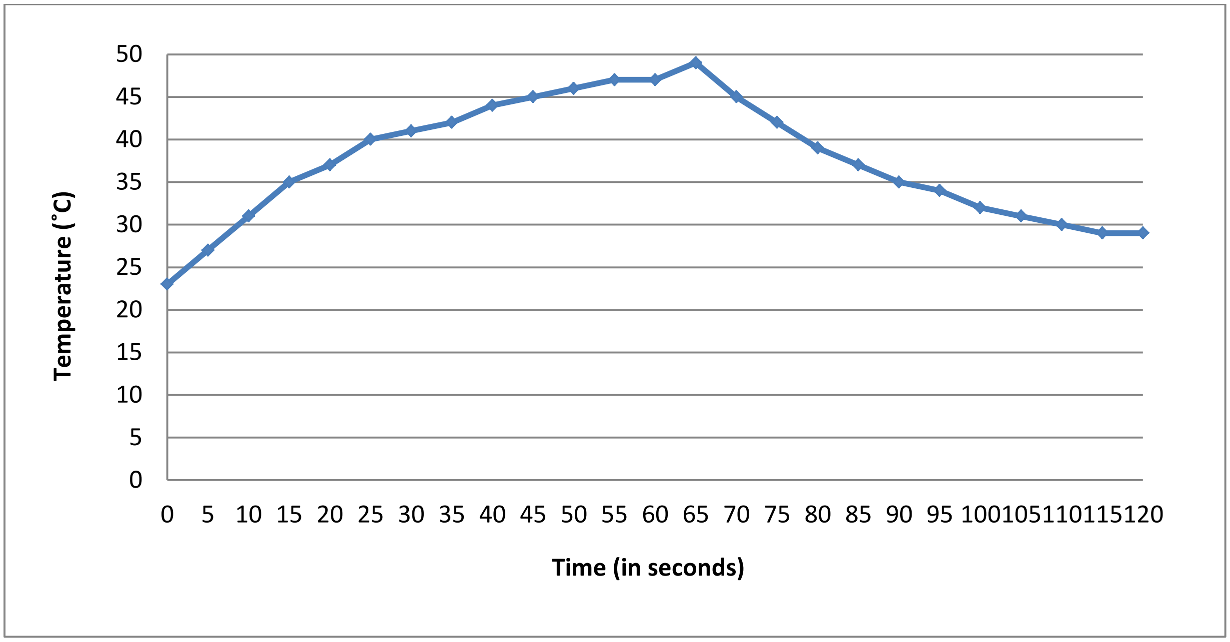

Figure 9 – heating/cooling curve of a dead bluet (large damselfly) arranged in a basking position.

The basking dead bluet (large damselfly), family Coenagrionidae, showed rapid heating, from 23˚C to 49˚C in 60 seconds under the heating lamp. The specimen showed very rapid cooling as well, cooling 15˚C in 55 seconds before the cooling curve leveled off, because of very small mass and characteristic dimension.

Note: small damselflies were too small to test using the probe in the laboratory

Δ T :

Table 1: Statistical analysis of field data of Δ T in different size and activity level categories of odonates. Asterisks denote sample categories used for Kruskal-Wallis analysis of median Δ T in figure 10 below.

Species Type	N	Mean	Std. Deviation	Median	Minimum	Maximum
	--------------------------------------ΔT °Celsius --------------------------------------					
Large Dragonfly Flight	2	10.0	1.4	10.0	9.0	11.0
Large Dragonfly Basking	1	3.0	-	3.0	3.0	3.0
Medium Dragonfly Flight*	30	9.3	3.7	9.0	.0	15.0
Medium Dragonfly Basking*	24	6.9	3.8	5.5	1.0	17.0
Small Dragonfly Flight	2	4.5	2.1	4.5	3.0	6.0
Small Dragonfly Basking*	12	6.2	2.7	6.5	2.0	9.0
Large Damselfly Flight*	4	5.3	4.0	4.0	2.0	11.0
Large Damselfly Basking	2	5.5	2.1	5.5	4.0	7.0
Small Damselfly Basking*	6	4.3	2.9	3.5	2.0	10.0
Aggregated Values	83	7.3	3.8	7.0	.0	17.0

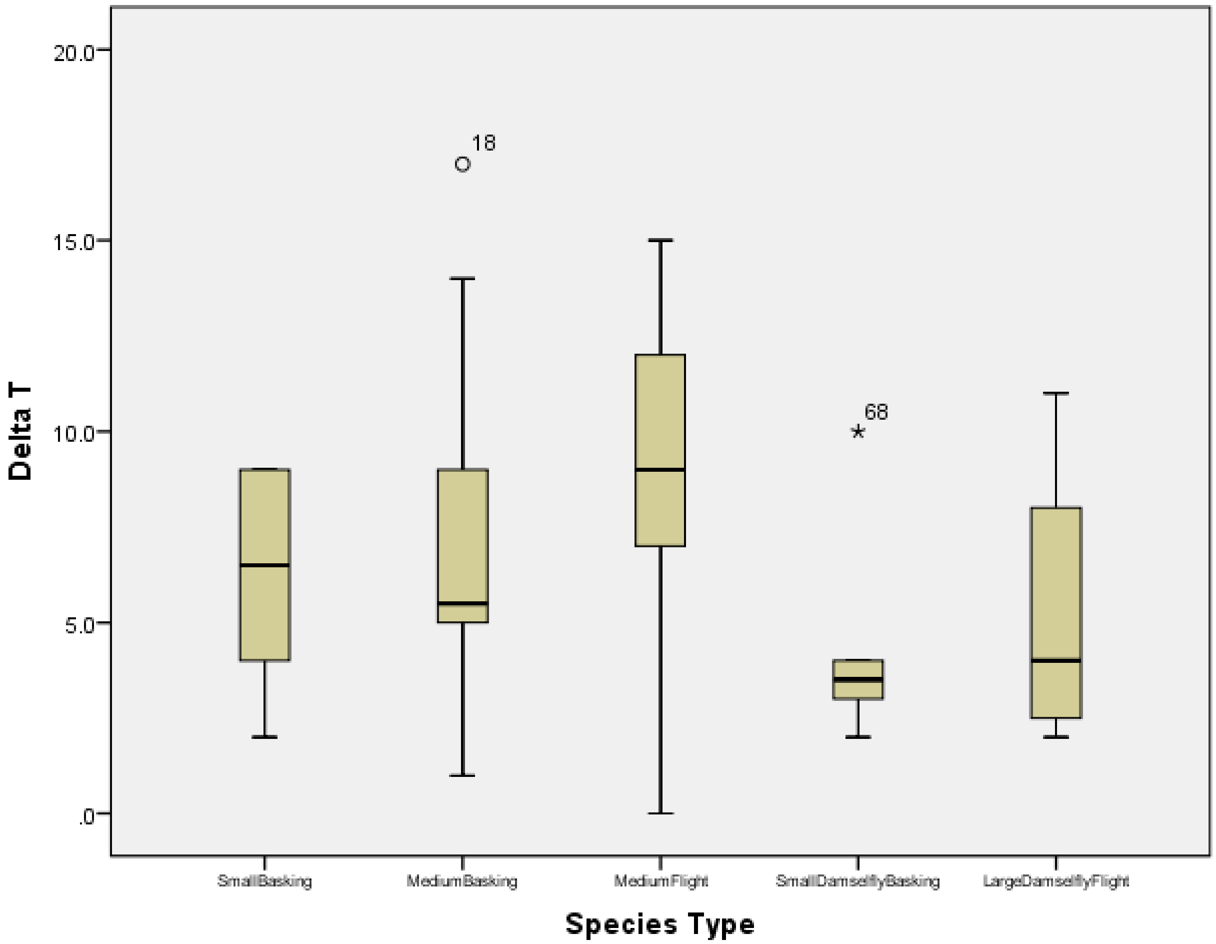

Figure 10 –Median Δ T values of varying size and activity level categories of odonates.

Of the 5 categories analyzed, medium dragonflies in flight had the greatest average and median Δ T and small basking damselflies had the lowest. The greatest range of variation in Δ T was seen in the large damselflies in flight, while the smallest range was found in the small basking damselflies.

Δ T was analyzed with a Kruskal-Wallis test using the median instead of the mean for each group (species type) under the null hypothesis that the medians of the 5 groups were the same. We obtained a Kruskal Wallis test statistic (adjusted for ties) of 15.8, with four degrees of freedom, and P = 0.003. This indicated that the median difference in temperature was significantly different for at least one pair of medians. We performed ten pairwise comparisons, using a Mann-Whitney U analysis which gave us an adjusted alpha of 0.005. Pairwise comparisons (which compare each median against every other median) only found one pair of species types that differed significantly: the medium dragonflies in flight and the small basking damselflies, which were the two extremes of the categories analyzed. For our comparison of small damselfly basking against medium flight, we obtained a p value of 0.002, which is significant at our adjusted 0.005 alpha level.

Determination of Variables Involved in Thermoregulation

1. **Haemolymph Flow From Wings to Thorax**

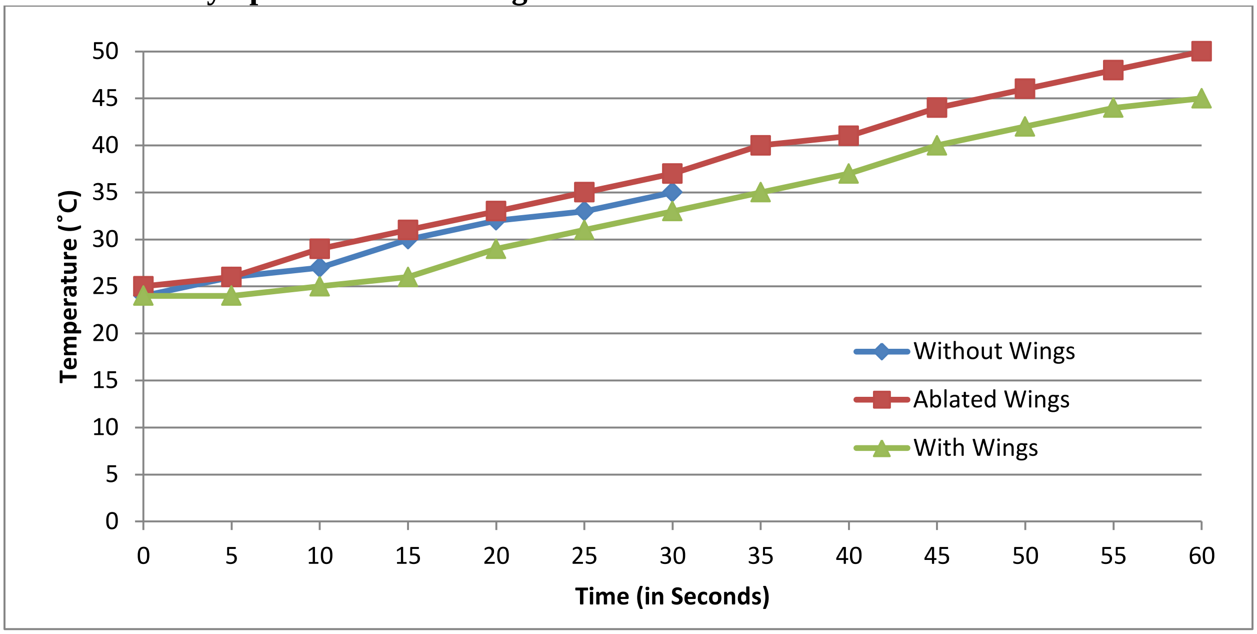

Figure 11 – comparison of the heating curves of a dorsally basking live blue dasher (*Pachydiplax longipennis*) with wings intact, with wings removed from body, and with wings ablated but reappressed touching the thorax.

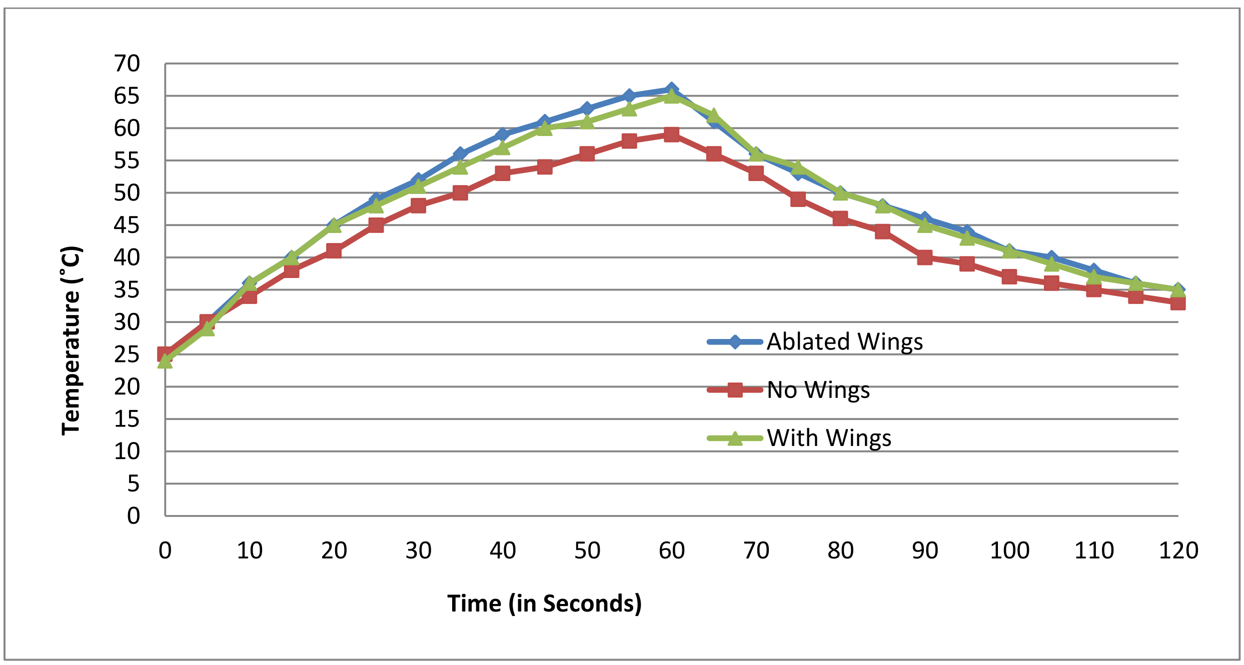

Figure 12 – comparison of the heating and cooling curves of a dead medium-sized odonate with wings, without wings, and with ablated wings in contact with the thorax.

The specimen with ablated wings in contact with the thorax did reach a slightly higher maximum temperature in the 60 second time interval, possibly due to reduced convective heat loss because of its greater characteristic dimension.

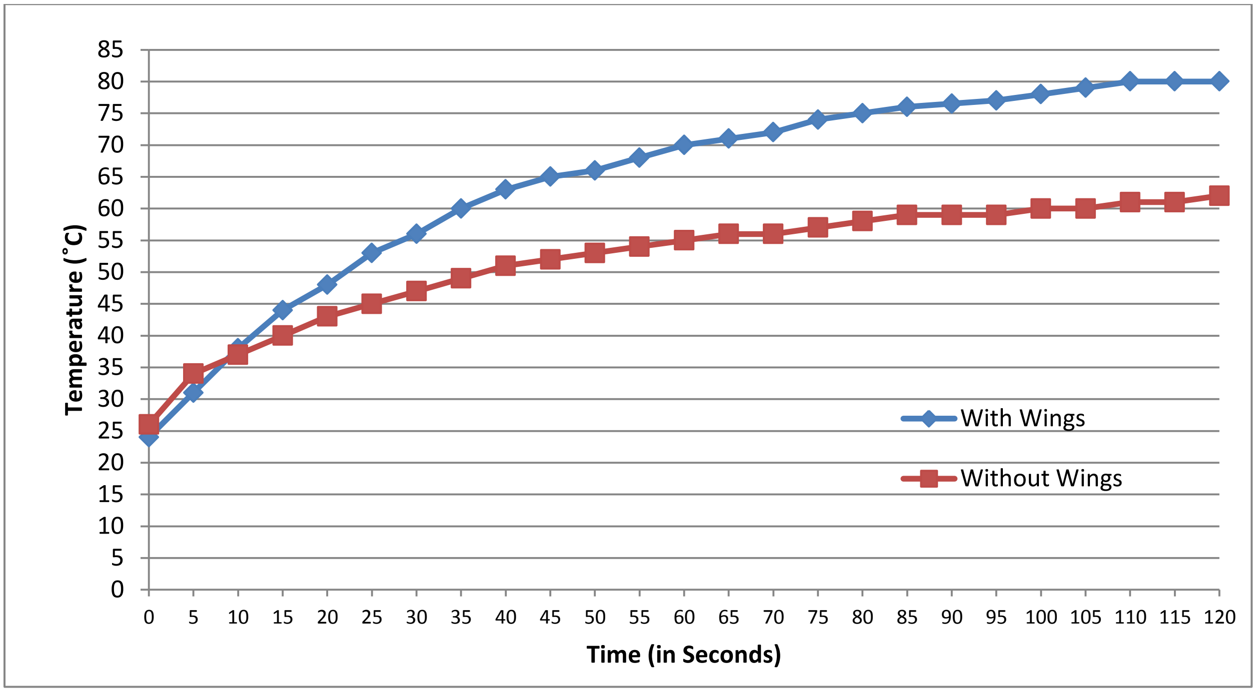

Figure 13 – comparison of the heating curves of dorsally basking dead blue darner (large dragonfly) specimens with wings and with wings mechanically separated from the thorax.

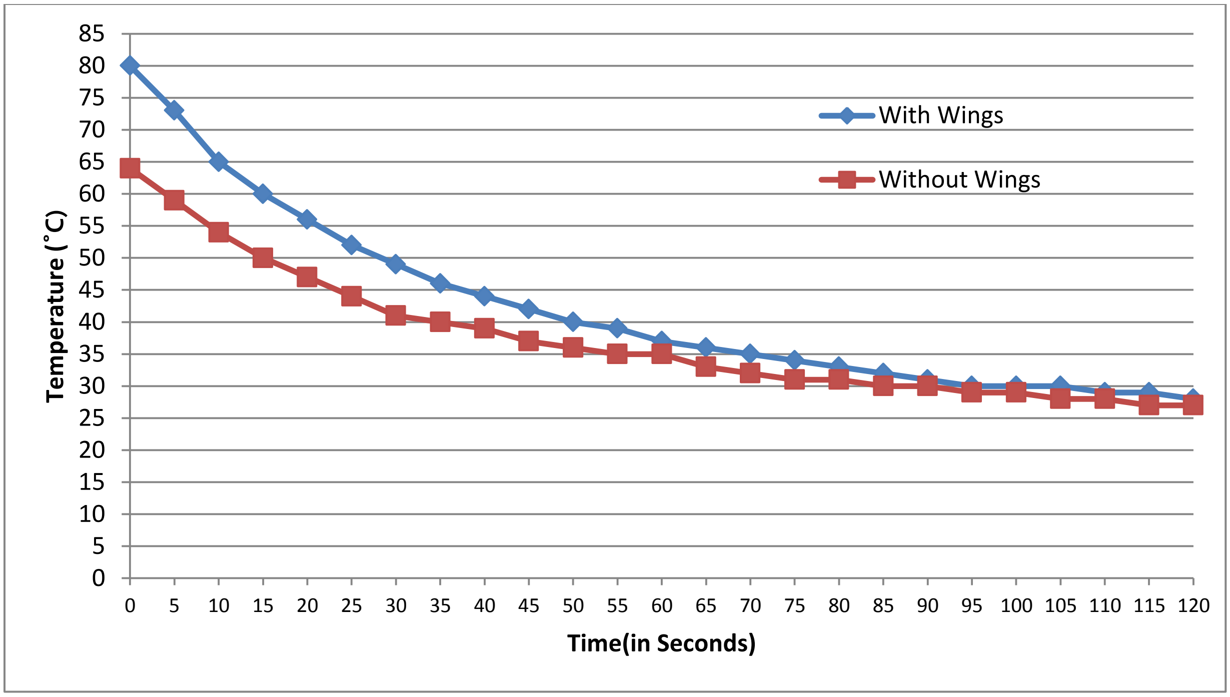

Figure 14 – comparison of the cooling curves of dorsally basking dead blue darner (large dragonfly) specimens with wings and with wings mechanically separated from the thorax

Figures 13 and 14 demonstrate the heating and cooling of a dead Blue Darner with and without wings. The majority of heating or cooling takes place in the first 60 seconds – after that, both curves level off.

Preferred Thermoregulatory Strategies by Odonate Size:

Small dragonflies only utilized passive strategies, particularly strategic basking positions, including obelisking. In medium dragonflies, passive and active strategies were both utilized. Passive strategies included varying dorsal basking positions and changing basking locations; no obelisking was observed. Frantic feeding and wing whirring were the active strategies utilized. Similarly, for large dragonflies, both passive and active strategies were utilized. They exhibited varying dorsal and vertical basking positions and selective basking locations. During warm weather, they would hang vertically (inverted obelisk) from vegetation. Wing whirring and water dipping were common active thermoregulatory strategies. Small damselflies were strictly passive and dependent on basking locations. Large damselflies utilized primarily passive thermoregulatory strategies and were mostly reliant on basking locations. Some wing "stretching" was demonstrated in some Jewelwings during warm weather.

Color Change:

No temperature-related color change was noted in any species of odonate.

DISCUSSION

I. Biodiversity Study

Forty-three species of dragonflies from five families and sixteen species of damselflies from three families were identified at PCCI between May 7th and August 10th 2012. This number is much greater than what we expected, due to several factors. First, Pierce Cedar Creek Institute comprises a large variety of different habitats, including prairies, woods, and wetlands. Second, there is a variety of aquatic environments, including Cedar Creek, Brewster Lake, small ponds and vernal ponds, to provide diverse aquatic breeding habitat. As a side note, 2012 became unseasonably warm early in the season, allowing migratory species to settle in Michigan earlier than usual, and also allowing larvae to metamorphose and emerge early in the season, leading to more possible breedings (for bivoltine species; species that experience two breeding seasons annually) in the year, which would lead to greater species density.

The number of damselflies at PCCI was very near the maximum number of damselflies recorded by the Michigan Checklist of Odonata (2004). The checklist had 18 species of damselflies identified in Barry County, of which we found 6 in addition to 10 unlisted species. However, the checklist only had 25 species of dragonflies recorded for Barry County (O'Brian 2004), and we found 17 of these species. However, we also found 26 unlisted species. No specimens from PCCI were recorded in the checklist, therefore, we can see that the odonate diversity of PCCI increases the previously recorded odonate diversity of Barry County.

II. Phenology Study

The Family Libellulidae was the most well-represented family of anisopterans at PCCI, exhibiting both the greatest diversity at 21 different species, and having one or more dominant (most abundant) species each week throughout the summer. Anisopterans were found mostly in prairies and adjacent woodlines, whereas the zygopterans at PCCI were found mostly in the woods, wooded creek areas, small clearings, or woodline edges. Odonates were abundant in early May, perhaps due to a very warm spring allowing for early migration of migratory species and early metamorphoses of aquatic larvae. There was a very large peak of species variety and abundance in late May through mid-June. High heat during the last week of June through mid-July, with record temperatures in the high thirties (C), caused populations to crash. The populations never recovered and the number of odonates at PCCI remained low throughout the rest of the summer.

III. Thermoregulatory Study

The effects of haemolymph flow from wings to thorax on thermoregulation:

We found that haemolymph flow from the wings to the thorax has a negligible effect on the thoracic temperature of odonates. Any effect that the wings have on thoracic temperature can be attributed to their biophysical mass increasing the characteristic dimension (diameter and length) of the odonate. The heating curve of the dead Ashy Clubtail is nearly identical when the wings are intact and when the wings are severed but still in contact with the thorax(reference heating curve figure here). However, when the wings are removed completely, the Ashy Clubtail both heated and cooled slower than when the wings were intact. There is a possibility that this is a result of the haemolymph leaking out of the individual's body, but specimens were not found to exhibit haemolymph leakage from wing stumps upon post-experimental examination. When the experiment was repeated with a live blue dasher, the results were similar. The Blue Dasher with the wings severed but still in contact with the thorax heated only slightly slower than when its wings were intact. The Dasher with the wings removed heated markedly slower than it did with intact wings. This is likely due to the effect of wings on reducing convective heat loss.

Statistical Analysis of Δ T

Our hypothesis that the average ΔT would increase with increasing thoracic size was only weakly supported by data collected under field conditions. When analyzing average ΔT we saw the largest Δ T in large dragonflies in flight (10 ˚C). Medium-sized dragonflies showed a 9.3 ˚C ΔT. Small dragonflies showed a ΔT of 6.2 ˚C. Unfortunately, no data was collected for small damselflies in flight as they spend the vast majority of their time basking, but field measurements showed a ΔT of 5.3 ˚C. However, when the median Δ T was analyzed using Kruskal-Wallis, we only found a significant difference between the medium dragonflies in flight and the small basking dragonflies. Due to small sample sizes, Δ T of large dragonflies was not able to be analyzed.

Preferred Thermoregulatory Techniques by Odonate Size

We originally hypothesized that larger odonates, such as the aeshnids and libellulids, would thermoregulate primarily through active strategies. As mass decreases, species should choose primarily passive means of thermoregulation. This hypothesis makes sense as one would expect larger dragonflies (with a larger muscle mass to surface area ratio than smaller dragonflies) to be able to produce a fair amount of metabolic heat through muscular thermogenesis and maintain an elevated thoracic temperature more easily than an insect with smaller muscle mass to surface area ratio, which would not be able to produce as much heat and would quickly return to ambient temperature via convective heat loss. Although larger dragonflies have a larger body volume to heat, they are also able to heat to a higher thoracic temperature than smaller dragonflies and exhibited a greater difference between thoracic and ambient temperature. Therefore, active thermoregulatory strategies would be more energetically sustainable and reasonable for larger odonates than for smaller specimens. Our hypothesis was weakly supported by the specimens observed and tested.

Large dragonflies exhibited a preference for active thermoregulatory strategies. They can produce a large amount of metabolic heat and retain it efficiently in their muscle mass. However, that ability can be disadvantageous at high ambient temperatures and high insolation, as active odonates can overheat. As a result, during the warmest part of summer months, aeshnids were mostly active after 6 p.m., when ambient temperatures and insolation began to drop. These factors combined to keep the individuals below maximum flight temperature. At various times and depending on ambient thermal conditions, large dragonflies were observed exhibiting wing whirring (a heating strategy) or water-dipping (a cooling strategy). During the hot afternoons, the large dragonflies could be found high in trees in the woods hanging vertically in the shade of the canopy. This strategy is an effective technique that reduces the amount of radiation absorbed from both the sun and ground vegetation and also allows the insect to cool by convective heat loss to the air and wind, helping to lower thoracic temperatures. Because representatives of Aeshnidae were the only dragonflies in the large size category, it is unknown as to whether this posture is unique to that family or if it is related to size.

The *medium-sized dragonflies*, mostly members of the families Libellulidae, Gomphidae, and Corduliidae, also followed the expected pattern of thermoregulatory strategies. They were observed to participate in both active and passive means of thermoregulation. Their active strategies included both wing whirring and frantic feeding. In addition, they were observed to bask in a variety of different locations in order to increase or decrease the amount of radiation they would absorb. However, no medium-sized specimens were observed exhibiting the vertical hanging posture utilized by the aeshnids. The medium species were also able to be active throughout a greater diurnal time period than the large dragonflies – from approximately 10 a.m. to 4 p.m.

The *small dragonfly* specimens were not observed participating in any active thermoregulatory strategies. Instead, they were completely dependent on passive means of thermoregulation: varying their basking postures and locations. Several species utilized a very interesting basking posture: the obelisk. This unique posture involves bending the abdomen at about an 80˚ upward angle from the thorax, and greatly reduces the amount of radiation being absorbed from the sun's rays. These dragonflies were active from around 10 a.m. through 4 p.m.

We hypothesized that damselflies would thermoregulate strictly by basking. The small species of damselflies did follow this pattern. However, the larger damselflies did not appear to be completely dependent on basking locations and time of peak activity to maintain their flight temperature: In July this hypothesized pattern was challenged by the activity of a few *Calopteryx maculata.* Some *Calopteryx* rested on low, shaded vegetation on the creek bank in what may have been an attempt to stay cool when ambient temperature exceeded 37 ˚C. Instead of holding still with the wings folded together above the back as is typical of heat avoidance behavior, individuals would very slowly lower both sets of their wings until they were extended dorsally out at the sides of the individuals at a 90˚angle, a position physiologically impossible for damselflies to maintain for an extended period of time during basking. Then, as the wings slowly reached a perpendicular position to the sides of the thorax, the insects would quickly pull their wings back up and clasp them together over their backs. We believe that this strategy is a form of active thermoregulation that could be utilized to help cool the insect through convective heat loss (by fanning the wings) without creating excess metabolic heat through the muscle activity that beating the wings might cause.

Small damselflies followed the expected strategies we hypothesized – they did not use either active means of thermoregulation nor did they alter their basking position: they were completely dependent on basking locations and time of activity. Consequently, they spent the majority of their time basking and seldom stayed in prolonged flight. Flight usually consisted of short stints from one basking location to another. In addition, they preferred to fly less than 1 meter above the ground vegetation. This makes intuitive sense, as the thoracic temperature of damselflies is closely aligned with ambient temperature, so they would benefit from flying near the horizontal boundary layer, where the temperature is more constant and less extreme, and convective heat loss is minimized in cooler weather. In addition, there is less extreme wind/air flow at the horizontal boundary layer near the ground.

Color Change: No physiological color change was noted in any species of odonate on the institute.

CONCLUSION

Our study showed that Pierce Cedar Creek Institute provides habitat to a much greater number and variety of odonates than expected, and while damselflies identified matched closely with the maximum number of species recorded in Barry County by the Checklist of Michigan Odonata (2004), the number of dragonflies at PCCI was nearly double the number of dragonflies recorded for Barry County (Obrien, 2004). Therefore, it is important to preserve a range of habitats such as woodlands, wetlands, and prairies in order to conserve the odonate biodiversity of a region. The diurnal phenology of the odonates varied by species, with smaller and medium dragonflies generally out earlier in the day and active into the afternoon, and larger dragonflies (such as the aeshnids) more active near dusk. Damselflies demonstrated a distinct preference for flying short distances and low altitudes, whereas the large dragonflies could fly as high as 10 meters and could remain in flight for extremely long periods of time. These are examples of how differently sized odonates can partition the thermal environment and alter diurnal phenology.

We noted some interesting results in the thermoregulatory study. As expected, most of the samples showed a greater mean Δ T when in flight than when basking, and Δ T decreased with decreasing mass. The heating and cooling curves were somewhat similar for differently sized odonates. We expected the larger odonates to heat and cool more slowly than smaller specimens; however, the curves were rather similar for large and small specimens. Because of their greater mass and lesser surface area to volume ratio, larger odonates were able to reach and maintain a greater thoracic temperature than were the smaller species. Haemolymph appeared to have no significant effect on the thermoregulation as shown in the heating and cooling curves for odonates with intact, separated, or completely removed wings. In fact, the wings themselves don't appear to play a significant part in thermoregulation – they are thin and transparent and therefore do not absorb a great amount of radiation. The extent to which wings can actually absorb and transfer energy to the thorax would be an interesting field for future study. Additionally, we saw that different sized odonates employed different thermoregulatory strategies. Large dragonflies used active (wing whirring and water-dipping) and passive (basking locations, time of activity, vertical hanging) strategies. Medium dragonflies also exhibited active (wing whirring and frantic feeding) and passive (basking position and location) strategies. Small dragonflies used only passive techniques, including the unique "obelisk" basking posture. Large and small damselflies relied strictly on activity times and basking location to thermoregulate. However, some large members of the family Calopterygidae participated in "wing stretching" from erect to dorsally extended wing positions.

ACKNOWLEDGEMENTS

My faculty mentor Dr. Matthew Douglas for his assistance throughout the course of the project.

Grand Valley State University Statistics Counseling Center, for assistance in statistical analysis and pairwise comparisons of thermoregulatory data (Delta T).

Pierce Cedar Creek Institute, for use of facilities and equipment throughout the course of the project. The Willard and Jessie Pierce Foundation and Undergraduate Research Grants for the Environment Program for funding the project

REFERENCES

Casey, Timothy M. *Insect Thermoregulation*. N.p.: John Wiley and Sons, Inc., 1981. 79-114. Print.

Casey, Timothy M., Jeri R. Hegel, and Charlene S. Buser. "Physiology and Energetics of Pre-Flight Warm-Up in the Eastern Tent Caterpillar Moth Malacosoma Americanum." *J. exp. Biol.* 18 Dec. (1980): 119-35. Print.

Conrad, K F., and G. Pritchard. "Female dimorphism and physiological colour change in the damselfly Argia vivida Hagen (Odonata: Coenagrionidae)." *Canadian Journal of Zoology* (1989): 298-304. Web. Accessed December 12, 2011.

Dubois, Robert. *Damselflies of the North Woods.* Duluth, MN: Kollath+Stensaas Publishing, 2009. Print.

Douglas, M.M. 1978. The Behavioral and Biophysical Strategies of Thermoregulation in Temperate Butterflies. PhD Thesis, University of Kansas, Lawrence, KS.

Gates, David M. *Biophysical Ecology*. Mineola, NY: Dover Publications, Inc., 2003. Print.

Gates, David M., and Rudolf B. Schmerl. Perspectives of Biophysical Ecology. 12th ed. New York: Springer-Verlag, 1975. Print.

Heinrich, Bernd. The Hot-Blooded Insects: Strategies and Mechanisms of Thermoregulation. Cambridge: Harvard University Press, 1993. 104-42. Print.

Heinrich, Bernd. Insect Thermoregulation. New York, Chichester, Brisbane, Toronto: John Wiley and Sons, 1981. Print.

Kirschbaum, K. 2007. "Anisoptera" (On-line), Animal Diversity Web. Accessed December 13, 2011.

May, Michael L. "Dependence of Flight Behavior and Heat Production on Air Temperature in the Green Darner Dragonfly *Anax Junius* (Odonata: Aeshnidae)." *The Journal of Experimental Biology* 24 July (1995): 23 85-92. Print.

May, Michael L. *Thermoregulation in Anisoptera*. N.p.: University of Florida, 1974. University of Florida. Web. 7 Oct. 2011.

Mead, Kurt. *Dragonflies of the North Woods*.2nd Edition. Duluth, MN: Kollath+Stensaas Publishing, 2009. Print.

Silbsy, Jill. *Dragonflies of the World*. Washington D.C.: Smithsonian Institution Press, 2001. Print.

Simpson, E. H. (1949) Measurement of diversity. Nature, 163, 688.

Stone, Graham N. "Endothermy in the Solitary Bee Anthophora." *Journal of Experimental Biology* 174 (1993): 299-320. Web. 9 Nov. 2012.

"Thermoregulation." *Encyclopedia of Insects*. Oxford: Elsevier Science & Technology, 2009. *Credo Reference*. Web. 14 December 2011.

O'Brian, Mark F. "Checklist of Michigan Odonata." *Umich.edu*. University of Michigan, 2004. Web. 13 Dec. 2011.

Walsberg, Glenn E., and Blair O. Wolf. "A Test of the Accuracy of Operative Temperature Thermometers for Studies of Small Ectotherms." *The Journal of Thermal Biology* 24 March 1996: 21, 275-281. Print.

Willmer, P G. "Thermal Constraints on Activity Patterns in Nectar-Feeding Insects." *Ecological Entomology* 8.4, November 1983: 455-69. Web. 9 Nov. 2012.

APPENDIX

Species List

Suborder Anisoptera:

Family Aeshnidae:

Aeshna Canadensis –	Canada Darner
Aeshna verticalis –	Green-striped Darner
Aeshna tuberculifera –	Black-tipped Darner
Aeshna umbrosa –	Shadow Darner
Aeshna constricta –	Lance-tipped Darner
Aeshna interrupta –	Variable Darner
Basiaeschna janata –	Springtime Darner
Anax junius –	Common Green Darner

Family Gomphidae:

Hagenius brevistylus –	Dragonhunter
Gomphus spicatus –	Dusky Clubtail
Gomphus lividus –	Ashy Clubtail
Gomphus exilis –	Lancet Clubtail
Gomphus fraternus –	Midland Clubtail
Dromogomphus spinosis –	Black-shouldered Spinyleg

Family Cordulegastridae:

Cordulegaster maculate –	Twin-spotted Spiketail
Cordulegaster bilineata –	Brown Spiketail

Family Macromiidae:

None sighted

Family Corduliidae:

Dorocordulia libera –	Racket-tailed Emerald
Epitheca cynosora –	Common Baskettail
Epitheca spinigera –	Spiny Baskettail
Epitheca canis –	Beaverpond Baskettail
Somatochlora williamsoni –	Williamson's Emerald

Family Libellulidae:

Libellula quadrimaculata –	Four-spotted Skimmer
Libellula incesta –	Slaty Skimmer
Libellula pulchella –	Twelve-spotted Skimmer
Plathemis lydia –	Common Whitetail
Libellula luctuosa –	Widow Skimmer
Libellula auripennis –	Golden-winged Skimmer
Sympetrum semicintum –	Band-winged Meadowhawk
Sympetrum internum –	Cherry-faced Meadowhawk
Sympetrum rubicundulum –	Ruby Meadowhawk
Sympetrumcostiferum –	Saffron-winged Meadowhawk
Sympetrum obstrusum –	White-faced Meadowhawk
Sympetrum vicinum –	Autumn Meadowhawk
Sympetrum corruptum –	Variegated Meadowhawk
Nannothermis bella –	Elfin Skimmer

Perithemis tenera –	Eastern Amberwing
Pachydiplax longipennis –	Blue Dasher
Erythemis simplicicollis –	Common Pondhawk
Pantala flavescens –	Wandering Glider
Leucorrhinia intacta –	Dot-tailed Whiteface
Leucorrhinia proxima –	Belted Whiteface
Celithemis eponina –	Halloween Pennant

Total Anisopterans: 43 individual species representative of 5 different families

Suborder Zygoptera:

Family Calopterygidae:

Calopteryx aequabilis –	River Jewelwing
Calopteryx maculata–	Ebony Jewelwing

Family Lestidae:

Lestes inaequalis –	Elegant Spreadwing
Lestes vigilax –	Swamp Spreadwing
Lestes rectangularis –	Slender Sreadwing
Lestes forcipatus –	Sweetflag Spreadwing

Family Coenagrionidae:

Argia moesta –	Powdered Dancer
Argia apicalis –	Blue-fronted Dancer
Enallagma signatum –	Orange Bluet
Enallagma exulans –	Stream Bluet
Enallagma carunculatum –	Tule Bluet
Enallagma civile –	Familiar Bluet
Enallagma erbium –	Marsh Bluet
Enallagma hageni –	Hagen’s Bluet
Ischnura verticalis –	Eastern Forktail
Nehalennia Irene –	Sedge Sprite

Total Zygopterans: 16 species representative of 3 families