Activity, Movements, and Microenvironment Associations of *Siren intermedia* (Lesser Siren) in a Western Kentucky Wetland Complex

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**Abstract** - In Kentucky, at the periphery of the species’ range, *Siren intermedia* (Lesser Siren) is a species of greatest conservation need. We monitored Lesser Sirens in a western Kentucky wetland complex from July 2018 to May 2019. Using funnel traps, we captured 60 Lesser Sirens and recaptured 2 individuals. Activity was greatest in July, and lowest October–January. Three Passive Integrated Transponder (PIT) telemetry surveys detected 7 PIT-tagged individuals; 1 Lesser Siren moved 36.9 m over 8 months. Capture success increased with warmer minimum water temperature. Body size upon first capture was unrelated to the microenvironment of the trap location. We recommend monitoring programs in western Kentucky concentrate funnel trapping in warm weather and use PIT telemetry to study movements.

**Introduction**

*Siren intermedia* Barnes (Lesser Siren) is a paedomorphic, aquatic salamander distributed in scattered populations throughout the eastern and central United States (Gehlbach and Kennedy 1978, Petranka 1998). The primary habitat of the Lesser Siren is shallow wetlands with permanent or semi-permanent hydroperiods that include dense aquatic vegetation and deep, organic sediment (Gehlbach and Kennedy 1978). Aquatic vegetation can provide nesting sites, foraging grounds (Gehlbach and Kennedy 1978), and cover (Brodman 2008). Lesser Sirens are nocturnal bottom-feeders that spend a considerable amount of time burrowed in deep, mucky sediment (Davis and Knapp 1953) where they estivate during dry conditions (Gehlbach et al. 1973, Reno et al. 1972). The habits of and habitats used by the Lesser Siren necessitate intensive and specialized efforts to capture individuals (Gehlbach and Kennedy 1978, Raymond 1991, Thompson et al. 2019). Thus, many aspects of the natural history of the Lesser Siren are poorly understood.

Specifically, few studies have investigated seasonal activity, movements, and microenvironment associations of the Lesser Siren. Previous research suggests that activity levels vary geographically. In the southern US, individuals appear to be active throughout the year during favorable environmental conditions (Frese et al. 2003, Gehlbach and Kennedy 1978, Godley 1983, Johnson and Blackwell 2011, Raymond 1991, Sever et al. 1996). In contrast, the Lesser Siren undergoes dormancy during cold winter months at the northern reaches of the species’ range (Blatchley 1899, Brodman 2008, Cagle 1942, Cagle and Smith 1939, Cockrum

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Manuscript Editor: Peter Paton
Even during times of activity, the Lesser Siren is considered relatively sedentary. For example, Gehlbach and Kennedy (1978) studied movements in a Texas population and found the maximum distance moved between captures was <12 m and larger individuals moved greater distances than smaller individuals. Movement distances and activity may be related to the microenvironment including organic sediment depth, water depth, and water temperature. Additional research is needed to evaluate activity, movements, and microenvironment associations of Lesser Sirens, especially on the edge of their range, where they are uncommon and populations are presumed to be highly disjunct.

In Kentucky, the distribution of Lesser Sirens is limited to 20 counties, and they are considered a species of greatest conservation need, threatened by habitat loss, habitat degradation and pollution (KDFWR 2013). The western Kentucky population is presumed to be highly disjunct due to its location on the range periphery (Brown 1984); the Ohio River separates western Kentucky from the northernmost populations in Indiana and Illinois, and the Mississippi River lies between western Kentucky and populations in Missouri. Baseline ecological information about the Lesser Siren is needed but lacking for Kentucky populations. In this observational study, we monitored the monthly activity, movements, and microenvironment associations of Lesser Sirens in a western Kentucky wetland complex.

Field-Site Description

Our study area was a wetland complex located in West Kentucky Wildlife Management Area (WKWMA), in McCracken County, KY (37°07’47.5”N 88°48’04.8”W), on the southern bank of the Ohio River in the Mississippi Embayment physiographic province (Fig. 1). The wetland had 4 dynamic, permanently inundated bodies of water with coarse woody debris from Castor canadensis Kuhl (American Beaver) activity. Based on aerial imagery (KyFromAbove 2013), we estimated the water surface area of our study area was 5519 m². Water surface area varied as inundation intermittently connected the north (water surface area: 2743 m²), south (1526 m²), and southeast pools (861 m²); the smallest pool (389 m²) was isolated by topography. Canopy cover of the terrestrial area was 90%, and the majority of the littoral and limnetic zone vegetation was Ludwigia sp. (water-primrose), Lemna minor L. (Common Duckweed), and Leersia oryzoides (L.) Sw. (Rice Cutgrass) (Price and Kreher 2016). There was a land fill east of the wetland complex, and a steep embankment contained the west side.

Methods

Data collection

In July 2018, we established 29 trapping stations, each with 2 funnel traps, around the perimeter of the 4 pools with 1 in the water channel connecting the north and south pools (Fig. 1). Due to accessibility concerns, trap stations were irregularly spaced; Euclidean distance between nearest neighboring stations averaged 14 ± 5 m (mean ± SD; min–max = 5–22 m). For 1 overnight period per month from
July 2018 to May 2019, we deployed these trapping stations and checked them the following morning. The interim between trapping sessions averaged $30 \pm 3$ d (min–max = 27–36 d), for a total study period of 301 days. We placed funnel traps on the surface of the water and submerged the funnel openings to ensure Lesser Sirens could swim inside, but by-catch would not drown (Frese et al. 2003). Water levels varied throughout the year; if a station was too shallow to submerge the funnel traps openings, then we did not deploy traps at the site. In the southeast pool, we set trap station 29 only if the other 3 trap stations were too shallow to set.

Figure 1. Placement of 29 funnel-trap stations (numbered in circles) for capturing Lesser Sirens in a wetland complex (white outline) at West Kentucky Wildlife Management Area, July 2018–May 2019 (KyFromAbove 2013).
Northeastern Naturalist
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We checked traps during the morning hours. Captured Lesser Sirens were measured, weighed, and uniquely marked by injecting each with a 134.2-kHz, 12.5-mm PIT tag (Biomark HPT12) using a Biomark© MK10 Implanter (Boise, ID). PIT tags have a high retention rate in sirens (Luhring 2009). We anesthetized individuals in a solution of maximum, double-medicated Church and Dwight© Orajel® (Active ingredients: 20% benzocaine and 0.26% menthol; Ewing Township, NJ) for processing (Cecala et al. 2007). When the solution became weak, we added 0.5 g of Orajel as needed and closely monitored each individual. Solution concentrations varied from 0.5 to 2.0 g per L, and induction times varied from 3 to 25 minutes. We processed specimens and returned them to their respective capture locations the same day.

To augment detection rates and provide additional information on movements, we opportunistically conducted PIT telemetry surveys along the wetland’s perimeter using a Biomark© HPR Plus portable reader with a BP portable antenna (Biomark, Inc) and attempted to observe tagged individuals (Oldham et al. 2016). We conducted surveys on 3 occasions (13 Nov 2018, 19 Mar 2019, and 16 Apr 2019). The hand-held reader emitted a radio frequency signal through an attached portable antenna and could detect a PIT-tagged individual up to 34 cm away, assuming optimal tag orientation and no environmental noise (Connette and Semlitsch 2012, Oldham et al. 2016).

To characterize spatial and temporal variation in the microenvironment of trap stations, we measured depths and water temperatures. Beginning in Aug 2018, we measured total depth (cm) and organic sediment depth (cm) using a graduated PVC pipe at every trap station, and calculated water depth (cm) from the difference between total depth and sediment depth. Hourly water temperatures were recorded at 18 random trap stations using Embedded Data Systems© Thermochron iButton temperature loggers (DS1922T-F5#, Lawrenceburg, KY) following a stratified-random design by pool; we grouped the channel (trap station 28) with the North pool. We coated the temperature loggers with white plastic tool dip (Plasti-Dip International©, Circle Pines, MN) to prevent water damage and secured them to the interior of a funnel trap with fishing line (Anderson et al. 2015). We did not collect microenvironmental data in July 2018.

Analysis
We quantified monthly activity based on capture rates in funnel traps. To estimate movements, we calculated the time (days), Euclidean distance (m), and movement rate (m per day) between initial capture site and either the recapture site in funnel traps or by detections using PIT-telemetry.

We quantified the microenvironment with 7 variables: minimum, maximum, mean, and standard deviation (SD) of the sequence of hourly overnight water temperatures (°C), organic sediment depth (cm), water depth (cm), and total depth (cm). We included the 3 depth variables, because the Lesser Siren is known to burrow in sediment (Davis and Knapp 1953). We included the 4 water temperature summary statistics because activity (as measured by the number of individuals captured) in other parts of the species’ range was related to the extremes and variability of
overnight temperature (Gehlbach and Kennedy 1978). Temperature loggers were not deployed at every trap station every trap-night, so we interpolated nighttime water temperature summary statistics to trap stations without loggers as follows. First, we estimated time of sunset and sunrise at our study site on each trap night with the R package ‘maptools’ (Bivand and Lewin-Koh 2020) and truncated the hourly sequences of water temperatures to nighttime, because the Lesser Siren is nocturnal (Davis and Knapp 1953). Then we calculated water temperature summary statistics at each logged trap station, and we assigned the mean of each summary statistic across trap stations within a pool to the unlogged trap stations in the same pool. We estimated the performance of this interpolation method by the root mean squared error (RMSE) of leave-one-out cross-validation (LOOCV) with training groups blocked by trap-night and pool to account for temporal and spatial structure of our water temperature data (Roberts et al. 2017). We did not deploy temperature loggers in July 2018, so those station-nights were omitted from the following analyses.

We modelled the associations between the microenvironment and funnel-trap capture success in a generalized linear mixed-effects regression framework followed by model selection. The unit of analysis was a trap station-night. To select the fixed effects of the global model, we calculated Pearson’s correlation coefficients \( r \) among the 7 microenvironment variables and omitted those that were highly correlated \( r > 0.9 \). We centered and scaled the remaining microenvironment characteristics. The global model also included random intercepts for trap night and trap station. We fit a global logistic linear mixed-effects regression model for capture success using the ‘glmer’ function from R package ‘lme4’ (Bates et al. 2018) and then proceeded with model selection. We used the ‘dredge’ function from R package ‘MuMIn’ (Barton 2020) to rank the set of all possible sub-models, calculate Akaike’s information criterion corrected for small sample sizes (\( \text{AIC}_c \)), and calculate each model’s \( \Delta \text{AIC}_c \) from the model set’s minimum \( \text{AIC}_c \) value. From the dredge output, we derived the likelihood of each model given the data \( \exp[-0.5 \times \Delta \text{AIC}_c] \). We considered models with a likelihood \( \geq 0.125 \) given the data and without uninformative parameters to be competing top models (Burnham and Anderson 2002).

We modelled the associations between the microenvironment and the body size (SVL) of Lesser Sirens upon first capture in a linear mixed-effects regression framework followed by model selection. The unit of analysis was a trap station-night that successfully captured a salamander. To select the fixed effects of the global model, we calculated correlations among the 7 microenvironment variables and omitted those that were highly correlated \( r > 0.9 \). We centered and scaled the remaining microenvironment characteristics. The global model also included a random intercept for trap night. We fit a global linear mixed-effects regression model with the ‘lmer’ function from R package ‘lme4’ (Bates et al. 2018) and proceeded with the same model-selection process we used in the analysis of capture success. All statistical analyses were conducted in R version 4.0.3 (R Core Team 2020). We present mean ± SD of measured variables and confidence intervals of fitted estimates throughout the paper.
Results

We set an average of 27 ± 2 (min–max = 23–28) trap stations each trap-night for a total of 295 trap station-nights and 590 trap-nights over 11 months. The number of trap stations with temperature loggers averaged 17.6 ± 0.2 (min–max =16–18 trap stations). We captured 60 individual Lesser Sirens in funnel traps and caught more Lesser Sirens in warmer months than colder months (Fig. 2). We re-captured 3.4% of marked individuals in funnel traps, and detected 12.1% with PIT tag readers (n = 58) (Table 1). Over the course of our study year, our limited use of PIT telemetry (3 winter surveys) accounted for 80% of our recaptures, and it was the only source of detections when water temperatures were low in November and March.

We measured movements between funnel-trap station locations of initial captures, recaptures, and detections by PIT telemetry (Table 1). We could not visually confirm the PIT detections, but evidence of movement as we attempted to hand-capture individuals indicated we detected a live animal. The Euclidean distance travelled by an individual between successive observations varied from 0.0 to 34.5 m over 28 to 301 d. The furthest cumulative distance travelled by an individual was 36.9 m over 8 months (26.3 m from August 2018 to November 2018, plus 10.6 m from November 2018 to April 2019); that individual was relatively small (SVL = 150 mm). Body size negatively correlated with distance moved (r = -0.375) and movement rate (r = -0.478).

![Graph showing monthly number of detections of Lesser Siren by funnel trap and PIT-tag telemetry survey](https://bioone.org/journals/Northeastern-Naturalist)

Figure 2. Monthly number of detections of Lesser Siren by funnel trap (grey bar) and PIT-tag telemetry survey (white bar) plotted against minimum underwater temperature averaged across trap stations (mean ± SD; black dashed line and gray ribbon) as measured by Embedded Data Systems© Thermochron iButton temperature loggers (DS1922T-F5#, Lawrenceburg, KY). Surveys were conducted along the perimeter of a wetland complex at West Kentucky Wildlife Management Area, July 2018–May 2019.
The microenvironment on a trap station-night was relatively stable throughout the study period with regards to organic sediment depth (22 ± 19 cm; min–max = 0–98 cm), water depth (18 ± 6 cm; 3–38 cm), and total depth (39 ± 18 cm; 14–101 cm), but we observed seasonal fluctuations in water temperature minima (10.6 ± 6.0 °C; 2.5–24.5 °C), maxima (14.1 ± 7.5 °C; 3.5–33 °C), means (12.4 ± 6.7 °C; 3.0–27.6 °C), and SD (1.1 ± 0.7 °C; 0.0–3.0 °C). The lower limits of water temperature minima and maxima occurred in Jan 2019, and the upper limits occurred in Aug 2018. The lower limits of water temperature SD occurred in Dec 2018 and Jan 2019, and the upper limits in Aug 2018 and Mar 2019. The performance of mean-interpolation was good for water temperature minimum (RMSE = 1.1), maximum (RMSE = 1.3), mean (RMSE = 1.0), and SD (RMSE = 0.3).

We assessed the association between the microenvironment and funnel-trap capture success on 266 trap station-nights, of which 35 successfully captured Lesser Sirens. (We omitted 28 trap station-nights in July 2018 and 1 in Sept 2018 that did not have microenvironmental data.) Water temperature minimum was highly correlated with water temperature mean ($r = 0.992$) and maximum ($r = 0.996$), and total depth was highly correlated with organic sediment depth ($r = 0.950$). After omitting highly correlated variables, the fixed effects of the global capture model were water temperature minimum and SD, organic sediment depth, and water depth; the random intercepts were trap night (10 levels) and trap station (29 levels). From a set of 16 sub-models, we found 7 with a likelihood $\geq 0.125$ given the data, 1 of which had no uninformative parameters. The top model ($\text{AIC}_c = 177.9$) indicated capture probability improved with increasing minimum water temperature (odds ratio = 3.54, 95% CI: [1.78, 7.05], $P < 0.001$). We did not capture Lesser Sirens on trap station-nights with minimum water temperatures below 6.5 °C.

We explored associations between the microenvironment and individual size upon initial capture. Mean SVL was 190 ± 38 mm ($n = 60$), mean TL was 285 ± 54 mm ($n = 60$), and mean mass was 37.7 ± 21.3 g ($n = 58$); 2 individuals >100 g

Table 1. Movement distances (meters), time elapsed (days), and movement rates (m per day) between detections of Lesser Sirens by funnel trap and passive integrated transponder (PIT) telemetry surveys along the perimeter of a wetland complex at West Kentucky Wildlife Management Area, July 2018–May 2019.

<table>
<thead>
<tr>
<th>PIT</th>
<th>Initial capture</th>
<th>Recapture</th>
<th>Movement</th>
</tr>
</thead>
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<tr>
<td></td>
<td>Date</td>
<td>SVL (mm)</td>
<td>Trap</td>
</tr>
<tr>
<td>989-35683</td>
<td>17 Jul 2018</td>
<td>203</td>
<td>4</td>
</tr>
<tr>
<td>989-35656</td>
<td>17 Jul 2018</td>
<td>275</td>
<td>27</td>
</tr>
<tr>
<td>989-35660</td>
<td>17 Jul 2018</td>
<td>195</td>
<td>14</td>
</tr>
<tr>
<td>989-35711</td>
<td>17 Jul 2018</td>
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<td>21</td>
</tr>
<tr>
<td>989-35678</td>
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<td>144</td>
<td>1</td>
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<td>142</td>
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</tr>
<tr>
<td>989-70705</td>
<td>13 Dec 2018</td>
<td>144</td>
<td>7</td>
</tr>
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</table>
were too heavy for our scale. One individual grew 10 mm in 301 d (0.03 mm per d) between initial capture and recapture (Table 1). The SVLs and associated trap station-night microenvironments of 40 individuals were included in this analysis, while 20 captures from July 2018 lacked microenvironmental data and were omitted. Water temperature minimum was highly correlated with water temperature mean ($r = 0.996$) and maximum ($r = 0.981$), and total depth was highly correlated with organic sediment depth ($r = 0.960$). The fixed effects included in the global model were water temperature minimum and SD, organic sediment depth, and water depth; the random intercept was trap night (6 levels). We dropped the random intercept for trap station (20 levels) from the global model, because its inclusion resulted in a singular fit (Barr et al. 2013). From a set of 16 sub-models, 5 had a likelihood $\geq 0.125$ given the data, and 1 (the global model) had uninformative parameters. The 4 competing top models each had 3 of the 4 fixed effects (Table 2). Although no coefficient estimates were significant, SVL was weakly correlated with water depth ($r = 0.080$), and SVL was negatively correlated with warmer minimum water temperatures ($r = -0.279$).

In a post hoc examination of the association between the microenvironment and body size, we observed a curvilinear relationship between SVL and minimum water temperature (Fig. 3). To test the significance of this relationship, we fit a linear mixed-effects regression model (Bates et al. 2018) where the response was SVL, the

<table>
<thead>
<tr>
<th>Fixed effects (estimate ± SE)</th>
<th>Model 1</th>
<th>Model 2</th>
<th>Model 3</th>
<th>Model 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intercept</td>
<td>181.6 ± 8.6*</td>
<td>181.5 ± 8.5*</td>
<td>182.9 ± 8.4*</td>
<td>181.3 ± 9.0*</td>
</tr>
<tr>
<td>Minimum temperature</td>
<td>-5.5 ± 8.7</td>
<td>-9.4 ± 9.4</td>
<td>6.6 ± 5.8</td>
<td>-3.7 ± 9.9</td>
</tr>
<tr>
<td>Organic sediment depth</td>
<td>7.2 ± 5.8</td>
<td>5.5 ± 5.7</td>
<td>7.4 ± 6.0</td>
<td>3.6 ± 6.0</td>
</tr>
<tr>
<td>Water depth</td>
<td>6.3 ± 6.2</td>
<td>7.4 ± 6.0</td>
<td>3.6 ± 6.0</td>
<td>0.6 ± 6.4</td>
</tr>
<tr>
<td>Temperature SD</td>
<td>1.5 ± 6.4</td>
<td>-0.5 ± 5.7</td>
<td>-0.6 ± 6.4</td>
<td></td>
</tr>
</tbody>
</table>

Random effects

- $\sigma^2 = 896.11$
- $\tau_{00} = 238.95$
- ICC = 0.21
- $n = 6$

Model fit

- $-\log(L) = -182.181$
- $AIC_c = 378.908$
- $\Delta AIC_c = 0.000$
- $\text{Likelihood} = 1.000$
- $\text{Weight} = 0.373$
fixed effects were minimum water temperature and its square, and capture date was included as a random intercept. We found SVL had a concave-down relationship with the square of minimum water temperature (estimate ± SE = -18.0 ± 8.1, 95% CI: [-29.8, -3.87], \(P = 0.026\)). Had this model been included in the model-selection process above, it would have been ranked first (AIC\(_c\) = 378.0, ΔAIC\(_c\) = -0.9).

**Discussion**

Activity of Lesser Sirens in western Kentucky was low during October–April and higher during May–September. Like populations in the northern limits of the species’ range (Blatchley 1899, Cagle 1942, Cagle and Smith 1939, Cockrum 1941), Lesser Sirens in western Kentucky may be dormant in cold months. High-activity seasons in the south are low-activity seasons in the north (Raymond 1991), suggesting there is an optimal ambient temperature for activity at the water’s surface.

Previously, Lesser Sirens were recorded moving only 10–20 m in water (Frese et al. 2003, Gehlbach and Kennedy 1978) and up to 6 m over land (Davis and Knapp 1953), with larger individuals moving farther than smaller individuals (Gehlbach et al. 2003).

![Figure 3. Locally-estimated scatterplot smoothing (LOESS) of association between snout-vent length (SVL) of Lesser Sirens (\(n = 40\)) upon first capture and minimum water temperature (°C) at capture site as measured by Embedded Data Systems© Thermochron iButton temperature loggers in a wetland complex at West Kentucky Wildlife Management Area, July 2018–May 2019.](https://bioone.org/journals/Northeastern-Naturalist)
and Kennedy 1978). We found 1 individual moved 37 m from initial capture site, and movement distances and rates were negatively correlated with larger body sizes. This site fidelity exhibited by larger individuals might be evidence of territorial behavior in male Lesser Sirens, which are typically larger than non-territorial females (Reinhard 2014). Farther movements by smaller individuals (SVL <150 mm; Hampton 2009) may be evidence of juvenile-based dispersal (Semlitsch 2008). PIT telemetry surveys improved movement estimates, but investigating connections between space-use and life-history traits (e.g., growth rates) of Lesser Sirens requires capturing and measuring individuals in hand.

Funnel-trap capture success improved with increasing minimum water temperature, but it was unrelated to water depth and organic sediment depth at the trap station. Low funnel-trap capture rates in cold months were likely due to Lesser Siren inactivity near the water’s surface (Luhring et al. 2016) or winter dormancy (Blatchley 1899, Cagle 1942, Cagle and Smith 1939, Cockrum 1941) rather than emigration from the wetland complex, because we detected 7 PIT-tagged individuals with telemetry in November and March. Our overall funnel-trap capture rates may have been constrained by the density of Lesser Sirens in western Kentucky, our sparse trapping-grid study design, and their potential to avoid traps (Johnson and Blackwell 2011). We placed traps 5–22 m apart, but other field studies placed traps 1.5–5 m apart and concentrated trapping efforts on microhabitats (Frese et al. 2003, Gehlbach and Kennedy 1978). Funnel-trap capture rates could be improved by concentrating trapping efforts in warmer seasons when Lesser Sirens are more active. Then PIT telemetry surveys may increase detections during low-activity periods, address problems associated with recapturing trap-shy individuals, and supplement a sparse trapping grid.

Mean body size of Lesser Sirens in western Kentucky and the single growth rate we documented were similar to values reported for the species (Brodman 2008, Cagle and Smith 1939, Frese et al. 2003, Hampton 2009, Sawyer and Trauth 2011). The maximum mass in western Kentucky is likely larger than our records, as 2 of the longest individuals we captured (SVL = 275 and 293) were too heavy (>100 g) for our scale. Minimum body size in western Kentucky may be smaller than our records, as smaller individuals are more likely to escape from funnel traps (Luhring et al. 2016). Indeed, 2 of the smallest individuals we captured had dark band markings, possibly injuries from attempting to escape the funnel trap.

Although our model-selection analysis did not establish a significant relationship between trap-station microenvironment and the body size of active Lesser Sirens, a post hoc analysis of these data suggests a quadratic response in SVL to minimum water temperature. Larger ectotherms may select microenvironments with warmer, less-variable water temperatures than smaller individuals (Fitzgerald and Nelson 2011). Alternatively, the apparent quadratic relationship could be an artifact of same-aged cohorts congregating in similar habitat (Brodman 2008). The ecology of the Lesser Siren in western Kentucky has not been investigated until this study (KDFWR 2013), largely due to the species’ cryptic and benthic nature. Future field research interested in studying the spatial ecology of the Lesser
Siren should increase the density of the funnel-trapping grids, focus funnel trapping on nights with minimum water temperature greater than 6.5 °C, and bolster recapture rates using a combination of PIT telemetry and funnel trapping. This more efficient, two-pronged approach could improve estimates of this elusive species’ population size and distribution, as well as reveal more information on its life history and ecology, which are essential data for anticipating and measuring the extent of threats to the long-term survival of the Lesser Siren in western Kentucky.

Acknowledgments

We thank Tina Marshall, Marshall County High School (MCHS), MCHS Environmental Science and AP Physics students, and Shelby Cosby for assistance in the field. Steve Hampson from the University of Kentucky (UK) Kentucky Research Consortium for Energy and Environment (KRCEE) Center for Applied Energy Research provided logistical support. Tim Kreher (Kentucky Department of Fish and Wildlife Resources [KDFWR]) provided site management information. Research collection permits (SC1711110, SC1811095) were provided by KDFWR. Funding was provided by KRCEE and the United States Department of Energy Portsmouth Paducah Project Office, United States Department of Agriculture McIntire-Stennis Cooperative Forestry Research Program (accession number 1001968), and UK Department of Forestry and Natural Resources. This research was approved under University of Kentucky Institutional Animal Care and Use Committee protocol (2013-1073).

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