- 1 Movement and home range of the Sickle Darter (*Percina williamsi*) in the upper Emory River of Tennessee, USA.
- 2 (Hydrobiologia)
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- 11 Abstract: Understanding movement patterns and home range of rare species is challenging, especially aquatic fauna like fishes. The Sickle Darter Percina williamsi is a rare fish species endemic to the upper Tennessee River basin in 12 13 eastern Tennessee, southwestern Virginia, and western North Carolina (USA). It has been listed as threatened by the 14 states of Tennessee and Virginia and is being petitioned for federal listing under the United States Endangered 15 Species Act. Little is known about the movement and home range of this species. A total of 8 Sickle Darters from 16 the upper Emory River system were implanted with 8-mm PIT tags and released at the point of capture. The mean 17 ( $\pm$ SD) total length and weight of all fish PIT tagged was 70.1  $\pm$ 3.4 mm and 3.08  $\pm$ 1.4 g. Movement of individuals 18 was tracked every two weeks for 6 months (September-March) with a Biomark® HPR Plus reader and BP Plus 19 portable antenna. Associated environmental data were collected throughout the study. Mean total effort for all the 20 tracking events was  $70 \pm 39.4$  min. mean catch-per-effort was  $9.3 \pm 6.6$  (min/detection) and mean ( $\pm$ SE) detection was  $69.5 \pm 12$  %. Mean ( $\pm$ SD) distanced moved of all individuals throughout the study was  $7.1 \pm 4.5$  m. Best sub-sets 21 22 regressions modelling suggest that Sickle Darter movement is related to discharge (m/sec<sup>3</sup>) at multiple temporal 23 levels (1, 3, or 7-day). Home range for individuals varied in size. Median home range size was 157.5 (86.0-312.5) 24  $m^2$ ) and median (range) degree of overlap for estimated home range was 23.3 (6.2-34.0) %. The results from this 25 study suggest that Sickle Darters exhibit strong site fidelity except when discharge is extremely high. Therefore, 26 conservation measures that protect or attempt to reconnect fragmented habitats will need to factor in the low 27 dispersal ability of this species. 28 Keywords: Conservation, movement, rivers, ecology, modeling 29 Declarations 30
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#### Introduction

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53 Understanding movement patterns and home range of rare species is challenging, especially aquatic fauna 54 such as fishes. (Holden, 1978; Rodriguez, 2002). Movement of large-bodied sport fishes, like salmonids, has been 55 widely documented on multiple scales (Holden, 1978; Rodriguez, 2002), however within-habitat, within-reach and 56 among system movements of small-bodied rare species are poorly understood. There are multiple reasons why 57 species move, such as seasonal spawning migrations, short-term movement to minimize stress (e.g., movement to a 58 thermal refugium), locating forage, or movement to another reach in response to a habitat disturbance (e.g., flood) or 59 loss of resources (e.g., food, cover; Hall, 1972; Rodriguez, 2002). Understanding the movement of a rare species can 60 improve the efficacy of monitoring its population trajectory. In addition, movement studies allow researchers to 61 understand how individuals respond behaviorally to environmental change, and how they may utilize available 62 habitat at various spatiotemporal scales (Holden, 1978; Rodriguez, 2002; Baxter, 2015; Cooke et al., 2016; Baker et 63 al., 2017; Pennock et al., 2018). This is important when determining conservation measures needed to preserve rare 64 species (Cooke et al. 2015). Movement studies on rare species can help determine critical habitat requirements 65 (Cathcart et al., 2017), which is important when considering that many freshwater species (~700) are considered 66 imperiled in North America as of 2008 (Jelks et al., 2008). Studies on fish movement allow for estimates of home 67 range in a particular system or habitat (Hill & Grossman, 1987). The size of a fish's home range is dependent on 68 multiple factors, such as life-history, biotic interactions, and abiotic factors. Fish size (total length) has been found to 69 have a positive relationship with home range size. This relationship has been observed in large-bodied species like 70 Largemouth Bass Micropterus salmoides (Lacépède, 1802) and small-bodied species like the European Bullhead 71 Cottus gobio (Linnaeus, 1758; (Minns, 1995). However, this relationship has not been observed within the family 72 Percidae, which include many imperiled, small-bodied darter species and larger-bodied common species like 73 Walleye Sander vitreus (Mitchill, 1818; Minns, 1995). Minns (1995) surmised that fish home range size was linked 74 to the metabolic activity of a fish, suggesting that larger fish have greater energetic demand. Consequently, these 75 fish will move greater distances to locate sufficient prey or refugia. This is important when considering the 76 conservation of small-bodied, imperiled fish species like darters, because species with small home ranges will be 77 less likely to disperse and colonize new habitat patches. Thus, population extirpation is more likely to occur when 78 their local habitat becomes unsuitable.

79 There have been many recent advancements in the applications of telemetry to small-bodied fishes (<150 80 mm total length) to help assess movement patterns and home range (Ruetzet al., 2006; Knaepkens et al., 2007; Baxter, 2015). Passive integrated transponder (PIT) tags have been used for decades to track many fish species from 81 all types of environments (Smyth & Nebel, 2013; Baxter, 2015). However, most species that are PIT tagged are 82 83 prized commercially or recreationally, are species that are easily recognized and valued by society (e.g., sharks) or 84 they are invasive (e.g., carps in North America). Recent telemetry studies have used PIT tags to track small-bodied 85 streamfishes that tend to be rare or of conservation value (Baxter, 2015; Baker et al., 2017; Cary et al., 2017; Kelly et al., 2017; Allan et al., 2018; Pennock et al., 2018). These researchers have outlined methods to track the 86 87 movement of individuals at large and small scales, and they have observed movement of individuals across multiple 88 habitat types within a stream.

The Sickle Darter *Percina williamsi* (Page & Near, 2007) is one rare fish species that has been
 understudied until recently (Jett 2010; Tennessee Wildlife Resources Agency [TWRA], 2015 Virginia Department

91 of Wildlife Resources [VDWR], 2015; Hecke & Alford, 2021). Historically, its distribution included the upper 92 Tennessee River basin (UTRB) in the states of North Carolina (NC), Tennessee (TN), and Virginia (VA; Etnier & 93 Starnes, 1993; Jenkins & Burkhead, 1994; Page & Near, 2007; Jett, 2010; Burns et al., 2007; TWRA, 2015; VDWR, 94 2015; Tracy et al., 2020; Hecke & Alford, 2021). Without a more complete understanding of this species, including 95 its movement and habitat usage, it is hard to prescribe suitable conservation measures to preserve it. At the 96 microhabitat scale, the Sickle Darter occupies flow-adjacent pools over a mix of substrate types (e.g., cobble, 97 boulder, sand, gravel, silt), and it is strongly associated with small woody debris or macrophyte cover. This species 98 is thought to remain in the same reach for most of the year, and individuals are captured in the same microhabitats 99 year after year (same range of depths, velocities, etc.). However, there have been cases where it moves to deeper 100 pools in the winter season (Etnier & Starnes, 1993). There is an ecdotal evidence that suggests they migrate short distances from pools to gravel areas of riffles for spawning, however no study has documented this (Etnier & 101 Starnes, 1993; J.R. Shute, personal communication). Studies on darter movement, in general, show that movement 102 103 tends to be species-specific and location dependent (Roberts & Angermeier, 2007; Roberts et al., 2008; Baxter, 104 2015). For example, Baxter (2015) found that Kentucky Arrow Darters Etheostoma spilotum (Gilbert, 1887) in 105 tributaries of the Red Bird River (Kentucky, USA) will move both upstream and downstream and cover distances 106 from 40 m - 4,000 m.

107 The goal of our study was to assess how the Sickle Darter moves spatially within a stream and to determine 108 temporal variation in its movement. We achieved this goal with the following objectives: 1) determine the 109 movement extent of the Sickle Darter in the upper Emory River system and the potential environmental drivers of 110 this movement, 2) assess the spatiotemporal variation in movement, and 3) determine the species' home range. This 111 study will further our knowledge of Sickle Darters by documenting how this species moves within its range, and it 112 will help inform future conservation measures to preserve this species.

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### Methods

115 Study Area

The Emory River is a spring-fed tributary system of the upper Tennessee River watershed in east
 Tennessee (Etnier & Starnes, 1993; TDEC, 2002; Fig. 1). This river originates in Morgan County, and it flows
 southeasterly until it meets its confluence with the Clinch River in Roane County, Tennessee (Tennessee
 Department of Environmental Conservation [TDEC], 2002). The Emory River main stem is 74 km long and its basin
 drains an area of ~2,300 km<sup>2</sup> (TDEC, 2002). This basin flows through two different Level III ecoregions: the

121 Southwestern Appalachian Mountains and the Ridge and Valley (Omernik, 1997).

122 Fish Collection and Tagging

123 Sickle Darters were captured from known occurrence locations within the Emory River drainages on two 124 different dates (09/27/2019 and 11/16/2019; Page & Near, 2007; Jett, 2010). This river was chosen because it 125 supports one of only two robust populations remaining in its fragmented distribution (Page & Near, 2007; Hecke & 126 Alford 2021). Backpack electrofishing and minnow seines (Bonar et al., 2009) were used to capture individuals. A total of eight Sickle Darters varying in size from 56 to 88 mm total length were collected, tagged, and released at 127 their point of capture in the Emory River system (two sites), which included (1) Rock Creek (width of  $\sim 12$  m, depth 128 129 of  $\sim 1$  m), a small tributary to the Emory River in Morgan County, Tennessee, and (2) the main stem upper Emory 130 River (width of  $\sim 10$  m, depth of  $\sim 1$  m) in Morgan County, Tennessee.

131 PIT tags were used to track individual Sickle Darter movement (Smyth & Nebel, 2013). The model of PIT 132 tags deployed were Biomark® HPT8 minichipTM (8.4mm X 1.4mm, 134.2KHz). These PIT tags are not known to 133 hinder growth, movement, or behavior of small benthic fishes (Ruetzet al., 2006; Knaepkens et al., 2007). Tagging methods closely followed Baxter (2015). Sickle Darters were tagged on the ventral side and on the posterior end 134 135 between the gular area and the vent. This area is the standard PIT-tagging location for small-bodied freshwater fishes (Kuechle & Kuechle, 2012; Baxter, 2015). A scalpel was used to make a small insertion at this location, then 136 137 the PIT tag was inserted by hand following the mid-ventral line at an approximate 45 angle. After insertion of the PIT tag, the location was treated with a petroleum jelly made of an antiseptic betadine solution. All materials used 138 139 were sterilized with 75% ethanol, and the individuals assisting with the PIT tagging of a fish wore nitrile gloves to

- 140 avoid potential infection of the PIT-tagging location. After insertion, each fish was checked for a unique PIT-tag
- 141 number. Fish were placed in a container of ambient river water with aeration and allowed to recover for 45 min.
- 142 After the recovery period, the tagged fish were released back to its capture location. Each individually tagged fish
- 143 was checked once again in the river for a corresponding PIT-tag number. PIT-tagging mortality and retention were
- assessed for tagged individuals throughout the study

## 145 Fish Tracking

146 The movement of PIT-tagged individuals was tracked biweekly after the original tagging date (09/27/2019;147 n=4) for 6 months (September-March). A second tagging date took place on 11/16/2019 (n=4). These PIT tags can 148 remain active for up to 70 years. Tagged fish were tracked using a Biomark® HPR Plus reader and a BP Plus 149 portable antenna. The antenna allows the PIT-tag reader to detect the tags under water, even if the fish is hiding 150 under cover (e.g., a rock or vegetation) simply by holding the reader approximately 30 cm from the animal. The 151 antenna has been found to sufficiently detect a benthic PIT-tagged species, the Mottled Sculpin Cottus bairdii 152 (Cope, 1872), which is strongly associated with rock cover (Kelly et al., 2017). Cross-channel paths (i.e., left bank 153 to right bank) were conducted in a zig-zag motion across the wetted width of the streamat each tagging site to track 154 the PIT-tagged individuals. These paths were done continuously until all fish were accounted for, or the detection 155 reach was covered ( $\approx$  500 m) at each tagging site. The paths did not overlap and there was <0.15 m between each of 156 the individual paths. Each time a PIT-tagged fish was located a weighted fluorescent marker was placed to identify 157 the point of detection. To determine if the "detection" was from a live fish, we used visual confirmation to determine 158 that the PIT-tagged fish was still alive (i.e., gill or body movement observed) and that the PIT tag had not been lost. 159 The corresponding geolocation of the "detected" Sickle Darter was recorded. The detection locations were marked 160 so that microhabitat and environmental data could be collected for each detected individual.

### 161 Environmental and Habitat Variables

162 Microhabitat characteristics were measured within a 2-m<sup>2</sup> area around the weighted marker (Table 1). These data included canopy cover (%), dissolved oxygen (mg/L), pH, stream depth (m), stream wetted width (m), 163 164 water temperature (C), water velocity (cm/sec), and percentage of substrate types (e.g., gravel, sand). Dissolved 165 oxygen and water temperature data were collected with a Pro20 Dissolved Oxygen Meter. The pH data were 166 collected with an Oakton PCSTestr 35 pH tester. Stream depth data were collected with a Keson 50-m field-167 measuring tape. Water velocity data (cm/sec) were collected using the neutrally-buoyant object method, whereby a 168 floating perforated plastic ball was timed as it drifted the 2-m distance at the area of detection (distance 169 traveled/time). This was done three times total to get an estimate of mean water velocity for the area of detection. 170 Substrate data were collected by visually determining the percentage of each substrate (sand/silt, gravel, cobble, and 171 boulder) at each detection location within the 2-m<sup>2</sup> detection area. Other environmental data were collected daily throughout the study for the Emory River watershed, and these data included discharge  $(m^3/sec)$  from the U.S. 172 173 Geological Survey (USGS) gauge # 03540500 at Oakdale, TN, precipitation (cm), and photoperiod (hours) from the National Oceanic and Atmospheric Administration (NOAA) National Weather Service (2020) climate station 174 175 GHCND: USW 00053868 at Oak Ridge, TN. Water temperature (C) data were collected every hour using two Onset 176 HOBO temperature loggers, with one deployed at the Rock Creek site and another deployed at the upper Emory River site. 177

# 178 Data Analyses

179 Sickle Darter movement data were analyzed in multiple ways. Movement was characterized by estimating 180 detection (0-1; Hubert and Fabrizo, 2007). Logistic regressions were run to assess the temporal relationship of 181 detection throughout the study. To determine the spatial movement of Sickle Darters, geolocations were plotted in ArcMap (ESRI, 2020), and the point-distance function was used to get an estimate of distance (m) between detection 182 points from tracking events. This was done for every tracking point for each tagged fish, such that distance moved 183 was determined by calculating distanced moved from the most previous tracking event. Mean movement distance 184 185 (±SD) was calculated for each tracking event. Furthermore, we determined the frequency of upstream and downstream movement throughout this study. A two-tailed Kolmogorov-Smirnov test was used to assess if 186 187 frequency of Sickle Darter movement upstream or downstream was distributed equally. We estimated a total 188 frequency of substrate use during each tracking event. We assessed the relationship of time on total frequency of

- 189 substrate with a simple linear regression. An ANOVA was run followed by a post-hoc Tukey test to determine if 190 significant differences of darter substrate-type use existed between tracking events. Statistical significance for all
- analyses were evaluated at an alpha = 0.05, and all analyses were completed in R (R Core Team, 2020; Zar, 1999).

192 We modeled the mean movement distance of Sickle Darters against the various temporal environmental 193 and microhabitat variables. We did this by using best-subsets regression modelling, a form of multiple regression 194 (Zar, 1999). We chose best-subsets regression modelling because the data were structured in a quantitative manner, that is, response and predictor variables were continuous. We also chose to use best-subsets regression modeling 195 196 because it is an efficient method to test all possible combinations of the predictor variables (MacNally, 2000). 197 Mallow's Cp and Adjusted R<sup>2</sup> were used to assess model fit at each temporal scale (7-day, 3-day, 1-day). We chose these temporal scales to capture delayed effects on Sickle Darter movement. Further, we used corrected Akaike 198 199 information criterion (AIC<sub>c</sub>) to determine the number of models to interpret at each spatial scale and the best model 200 in each model group. Corrected Akaike information criterion was used to account for the small samples size used in our analyses. At each spatial scale, all models with  $\Delta AIC_c$  value  $\leq 5$  were interpreted further (A kaike, 1973; 201 Burnham & Anderson, 2004; Symonds & Moussalli, 2011; Liao et al., 2018). We further interpreted our best models 202 203 at each spatial scale by assessing model fit with Analysis of Variance. To minimize effects of multicollinearity, 204 variables with VIF <4 were interpreted further in the analysis of variance.

205 To estimate the home range (90% of contour) and the core range (50% of contour) for each PIT-tagged 206 Sickle Darter, the kernel density tool was used in ArcMap (ESRI, 2020). The Fish Tracker tool in ArcMap was used to smooth out the home range estimates and make them fit the actual riverine system where this study took place 207 208 (upper Emory River watershed; Laffan & Taylor, 2013). This tool applies the home range estimate to a more fishlike habitat (rivers), by making the estimate fit the aquatic environment more, compared to estimates of home range 209 for terrestrial species (Laffan & Taylor, 2013). Total and median home range size (m<sup>2</sup>) for each PIT-tagged Sickle 210 211 Darter was estimated. A rea of home rage was estimated rather than the linear home range because the estimates of 212 home range were on such a small scale. We assessed the relationship of size (total length in mm) of PIT-tagged fish 213 on home range with simple linear regression modeling.

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Results

216A total of eight Sickle Darters were tagged on two different dates. At the Rock Creek site, six individuals217were tagged, and two individuals were tagged at the upper Emory River site. On the first tagging date (09/27/2019)218there was an initial tagging mortality rate of 25% (1 of 4 fish). One fish died after being tagged, but this fish was219small in comparison to other PIT-tagged fish (56 mm) and showed signs of stress immediately after capture and220prior to tagging. On the second date (11/16/2019), there was an initial tagging mortality rate of 0%. The mean (±SE)221size of all PIT-tagged fish was  $70(\pm 4.1)$  mm and  $3.1 (\pm 0.5)$  g.

222 A total of 10 tracking events were conducted. Our study was cut short due extreme water flows that seemed 223 to displace Sickle Darters outside of our detectable range or cause mortality. In February of 2020 the Emory River 224 experienced a record flood event (2089 m<sup>3</sup>/sec at the USGS Gauge at Oakdale, TN on 02/06/2020). Sickle Darter movement declined throughout out the study (linear regression, F=2.08, df=8, P-value = 0.03,  $R^2=0.21$ ; Fig. 2). Our 225 226 detection of Sickle Darters also declined significantly throughout the study (logistic regression,  $\beta$ =-31.92, odds 227 ratio=<0.01 P-value=<0.01). Like detection, this was likely caused by the high flow event. The frequency of Sickle Darter movement downstream or upstream from its capture site was not significantly different (D=0.19, P-value= 228 229 0.88; Fig. 3). There was no significant relationship between time and the four substrate types utilized at each detection location during each tracking event (sand: 1.28, df=7, P-value=0.30,  $R^2$ =0.18; cobble: 2.02, df=7, P-value=0.30, R^2 230 value=0.21,  $R^2 = 0.25$ ; boulder: 0.11, df=7, P-value=0.76,  $R^2 = 0.02$ ; gravel: 1.11, df=7, P-value=0.33,  $R^2 = 0.16$ ). 231 An ANOVA with post-hoc tukey test was not utilized because there were no significant relationships. Nevertheless, 232 233 sand and cobble were utilized the most throughout the study (Fig. 4).

There was little variation in the relationship between environmental variables and Sickle Darter movement across the three temporal scales. At the 1-day temporal scale, the top 5 best-subsets models associated with 236 movement included median daily discharge, precipitation, and daily temperature change, with the model including

median daily discharge being the best model (6.14 AIC<sub>c</sub>; Table 1). However, models that included median daily  $(7.02 \pm 1G)$  (7.02  $\pm 1G$ ) (7.02 \pm 1G) (7.02  $\pm 1G$ ) (7.02  $\pm 1G$ ) (7.02 \pm 1G) (7.02  $\pm 1G$ ) (7.02  $\pm 1G$ ) (7.02 \pm 1G) (7.02 \pm 1G) (7.02  $\pm 1G$ ) (7.02 \pm 1G) (7.02 \pm

discharge and precipitation (7.80 AIC<sub>c</sub>), daily temperature change (7.83 AIC<sub>c</sub>), and precipitation (7.90 AIC<sub>c</sub>) fit the
 data well. At the 3-day temporal scale, the top 5 best-subsets models included median daily discharge, daily

temperature change, mean daily temperature, and precipitation, with the model including median daily discharge

241 being the best model (6.14 AIC<sub>6</sub>: Table 1). However, models that included daily temperature change (5.95 AIC<sub>6</sub>).

242 mean water temperature (6.11 AIC<sub>c</sub>), and precipitation (6.16 AIC<sub>c</sub>) also fit the data well. At the 7-day temporal

scale, the top 5 best-subsets models included median daily discharge, daily temperature change, mean daily

temperature, and precipitation, with the model including median daily discharge being the best model (8.39 AICc
 Table 2). However, models that included median daily discharge and precipitation (8.44 AICc), and mean daily

245 rable 2). However, models that included mediandary discharge and precipitation (8.44 AIC<sub>c</sub>), and mean daily discharge, and precipitation (9.41 AIC<sub>c</sub>) also fit the data well. The top models from

each scale and predictor variables were retained for interpretation because they met criteria for further interpretation

analyses (Table 2). At the 1-day temporal scale, the median daily discharge was negatively associated with Sickle

249 Darter movement, but this relationship was not statistically significant (t=-1.88; P-value =0.16). At the 3-day 250 temporal scale, the median daily discharge was negatively associated with Sickle Darter movement, but this

relationship was not statistically significant (t = -1.82; P-value = 0.14). At the 7-day temporal scale, the median daily

discharge was negatively associated with Sickle Darter movement and was statistically significant (t = -6.51; P-

value =<0.01). No other model variables as ide from median daily discharge had a VIF <4, so they were not

considered further in the analysis of variance.

Sickle Darter home range size varied individually (Fig. 5). Only PIT-tagged fish from Rock Creek were
 considered for the home range analyses. The median (min.-max.) size of home ranges was 157.5 (86.0-312.5) m<sup>2</sup>.
 There was no significant relationship between PIT-tagged fish size and home range size (5.05, df=5,P-value = 0.09, R<sup>2</sup>=0.56; Fig. 6).

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#### Discussion

Sickle Darter movement varied temporally during our study, but overall, they moved very little from a 261 262 spatial context. Thus, it is likely that Sickle Darters exhibit high site fidelity in this river system, especially during average to low discharge. Prior to this study, there was anecdotal evidence suggesting that Sickle Darters in Little 263 264 River (Tennessee, USA) move to deep pools during the winter months and to shallow gravel riffles to spawn in the spring (Etnier & Starnes, 1993; J.R. Shute, personal communication). However, the movement of Sickle Darters in 265 266 the Emory River system may be different compared to that in the Little River. The Little River is considered a small 267 to medium-sized river, and it has a mosaic of heterogenous riverine features, such as riffles, runs, and pools with 268 highly variable depths and substrates. The upper Emory River system, on the other hand, consists of short and few riffles with shallow pools and runs (<1 m deep) and a relatively homogeneous mix of sand, silt, and cobble 269 270 substrates. The riverine features of these two systems, and amount of available habitat may influence the extent to 271 which individuals from these two fragmented populations move. Other studies on darter movement have found 272 different results pertaining to the distance moved by darters (Roberts & Angermeier, 2007; Roberts et al., 2008; Holt 273 et al., 2013; Baxter, 2015; Hicks & Servos, 2017). Roberts et al. (2008) found that the Roanoke Logperch, P. Rex, 274 exhibited high site fidelity throughout their tagging study. Holt et al. (2013) and Hicks & Servos (2017) found that the Brown Darter E. edwini (Hubbs & Cannon, 1935) and Rainbow Darter E. caeruleum (Storer, 1845), 275 276 respectively, exhibited high site fidelity throughout their tagging studies. In contrast, the Blackbanded Darter, P. 277 nigrofasciata (Agassiz, 1854), a species more ecologically and phylogenetically like the Sickle Darter, was found to move farther distances (max distance moved of 420 m) than what we report for the Sickle Darter (Freeman, 1995). 278 279 These differences may be due to the shifting sandy bottoms treams that Blackbanded Darters occupy in coastal plain 280 ecoregions, compared to Sickle Darters which are found in more interior mountain streams. Baxter (2015) found that 281 Kentucky Arrow Darters can move a large distance as well, with some individuals moving up to 4 km. Thus, differences in movement of darters are probably due to a multitude of factors, dependent on species and location 282 283 (i.e., stream type). Unfortunately, our study was cut short due record flooding in the Emory River, which resulted in 284 displacement of PIT-tagged individuals outside of our detectable range or caused mortality due to the high flow

event. This prevented us from observing Sickle Darter movement during the spawning season of this species (late

February to early April; Etnier & Starnes, 1993). A new movement study should be completed to observe how this

species moves on an annual basis to encompass the spawning season and summer months which we failed to

observe in our study. Further studies should also consider how the movement of this species potentially varies in
 other rivers within its range. There are three remaining viable populations of Sickle Darters (Hecke & Alford, 2021)

290 in the upper Emory River sub-basin, Little River sub-basin, and Middle Fork Holston River sub-basin. It is possible

that Sickle Darters move differently in these sub-basins due to each sub-basin's unique riverine features, size, and

- amount of available habitat (Ward, 1998). These populations are separated by dams and their impoundments.
- Because we found that Sickle Darters exhibit high site fidelity, it may be unlikely that these populations would mix
- because of dispersal.

295 Sickle Darter movement can be linked to changes in discharge. We found that discharge, no matter the temporal scale, had a negative influence on Sickle Darter movement. However, we did observe that discharge over 7 296 297 days prior to tracking appears to be more important than discharge for 3 days and 1 day prior to tracking. In 298 response to changes in discharge, Sickle Darters appeared to move less when there is increased variation in 299 discharge. Albanese et al. (2004) found that flood events can strongly affect the movement of small-bodied stream 300 fishes, which further supports our findings that Sickle Darter movement is linked to discharge. Other studies have 301 found that darter movement is related more to the amount of available habitat and multiple environmental 302 characteristics within a specific river. (Roberts & Angermeier, 2007; Roberts et al. 2008). Mundahl & Ingersoll 303 (1983) found that the Johnny Darter E. nigrum (Rafinesque, 1820) and Fantail Darter E. flabellare (Rafinesque, 304 1819) movement during fall months was driven by population density and quality of habitat. Baxter (2015) found 305 that there was very little seasonal effect on the movement of Kentucky Arrow Darters. We observed a significant 306 change in water temperature in our study, but this variable was not a significant driver of Sickle Darter movement. If 307 tracking could have been conducted for a full year, then water temperature may have been identified as an influential variable on Sickle Darter movement. We were only able to track Sickle Darters during fall and winter, when water 308 309 temperature may not be as important as during the spring spawning season. Future studies should look at the 310 potential relationship of Sickle Darter movement throughout a complete seasonal cycle to determine if water 311 temperature plays a significant role in the movement of this species.

312 Microhabitat utilized by Sickle Darters throughout this study remained constant. Sickle Darters were found 313 to inhabit the same substrate frequency at each detection site during each tracking event (i.e., sand and cobble). 314 Other studies on darter movement have found varying results. Skyfield et al. (2008) found sex-linked differences in 315 microhabitat use by the Gilt Darter P. evides (Jordan & Copeland, 1877). We did not distinguish between male and 316 female individuals in our study. Future studies should consider this component when looking at the movement of the 317 Sickle Darter, but males do not exhibit sexual dimorphism like most darter species, thus a sex-specific study would be challenging. Holt et al. (2013) found that Brown Darters did not move to different microhabitats, but rather 318 319 moved to different areas of the river that had the same available microhabitats. Baxter (2015) also found that 320 Kentucky Darters did not move between microhabitats, but rather moved to different areas where the preferred microhabitat was available. Freeman (1995) found that the Blackbanded Darter moved across different habitats to 321 322 reach a desired microhabitat. The section of Rock Creek where we observed Sickle Darter movement is not 323 comprised of a mosaic of habitats, and habitat is homogenous, consisting primarily of cobble and sand substrates 324 and shallow pools.

325 Sickle Darter (adult) home ranges are relatively small compared to many other freshwater fish species 326 (Minns, 1995). There have been very few home range studies on darters, but home range of small-bodied stream 327 fishes appears to be small (Gerking, 1953; Winn, 1958; Hill & Grossman, 1987; Rakocinski, 1988; Freeman, 1995; Minns, 1995; Hicks & Servos, 2017). Hicks & Servos (2017) found the Rainbow Darter E. caeruleum had a very 328 329 small home range (median = 5 m) and remained in the same riffle in which they were tagged. This is similar to our 330 results, where Sickle Darters had a small home range and were found in the same habitat type over time. Winn (1958) estimated the food, reproductive, and escape range (all of which comprise the home range) for 10 species of 331 darters in rivers and reservoirs, finding that home range was very small (< 5 m) for each species. However, these 332 333 estimates of home range were based off visual observations, and no tagging or mark-recapture study was conducted 334 to quantitatively determine home range. Scalet (1973) found that Orangebelly Darters E. radiosum (Moore &

Rigney, 1952) appear to have a small range but did not estimate actual size of this species' home range. Compared

to other benthic species, like the European Sculpin (45-m<sup>2</sup> home range) and the Banded Sculpin *C. carolinae* (Gill,

1861; 47 m<sup>2</sup>), the home range of the Sickle Darter is substantially bigger (Greenberg & Holtzman, 1987;

338 Downhower et al., 1990; Minns, 1995). This study outlines a method to estimate the home range of darters and other

- rare, benthic, and small-bodied fish species and it may also facilitate/inspire future tagging studies on imperiled
   small-bodied fishes. Future research should consider how Sickle Darter home ranges vary from sub-basin to sub-
- 340 small-0 341 basin.

342 Our study suggests that an interesting relationship exists between hydrology and Sickle Darter movement. 343 Future research should explore this relationship by assessing this species' critical swimming speed in the presence 344 and absence of refugia (habitat complexity; Scott & Magoulick, 2008). This will help biologists and researchers 345 understand what happens to the Sickle Darter during high flow events. Further, this will also help shed light on the functional organization of this species within the fish assemblage (Poff & Allan, 1995). With a more variable 346 347 environment (more frequent high flow events) being a likely result of climate change, understanding the hydrologic and climatic factors that negatively affect populations of Sickle Darters will be key to the preservation of this rare 348 fish (Ficke et al., 2007; Hecke & Alford, 2021). Future research should consider the movement of Sickle Darters on 349 350 a smaller temporal scale. We only assessed Sickle Darter movement every ~2 weeks between tracking events, this 351 may have caused us to underestimate how much Sickle Darters move. Future movement studies based on PIT tagging, should consider using flatbed (streambed) arrays to detect PIT-tagged fish, this would allow for fine scale 352 353 (daily) and more estimates of Sickle Darter movement, rather than the portable antenna that we used in this present 354 study (Johnston et al., 2009).

355 PIT tagging of rare, small-bodied fish like darters, is possible and yields a high PIT-tag retention and tagging-survival rate. This study outlines a way to conduct movement studies on similar small-bodied imperiled 356 357 fishes. We experienced a low tagging-mortality ( $\sim$ 14%), and tag loss (0%) throughout this study, which is supported 358 by other PIT-tagging studies on other small benthic fish species (Baxter, 2015; Knaepkens et al. 2017). Baxter (2015) observed similar results with tagging-mortality (none reported) and tag loss (0%) on the Kentucky Arrow 359 360 Darter, Ideally, we would have retained individuals outside of our actual study and monitored PIT-tag retention and mortality through a pilot study, but due to the rareness (proposed for federal listing; USFWS, 2011) of the Sickle 361 Darter, we were unable to collect a large number of fish to support such a study. Nonetheless, we did find that Sickk 362 363 Darters  $\geq$  55 mm can support PIT tags. This is supported by Baxter (2015), who suggest that larger individuals of a 364 darter species should be able to support PIT tags. This leads to higher tag survival and retention rates. Knaepkens et al. (2017) PIT-tagged European Bullheads (50-94 mm) and found relatively low tagging mortality (~10%), which 365 further supports the premise that larger specimens of small-bodied fish can be PIT-tagged. 366

367 Our study provides further knowledge to the understanding of Sickle Darters. Adding to our knowledge base of Sickle Darters will be important for the future of this species as it was proposed federal listing under the U.S. 368 Endangered Species Act(US, 1973; TWRA, 2015; VDWR, 2015; USFWS, 2011). This species is considered an 369 370 imperiled species due to anthropogenic factors in the upper Tennessee River basin, particularly habitat 371 fragmentation from dams and other environmental disturbances (Hampson et al., 2002; Jelks et al., 2008; Angermeier & Pinder, 2015; Hecke and Alford, 2021) This study developed further research questions for this 372 373 species which should be addressed when considering how to preserve the Sickle Darter. However, our study found that Sickle Darters exhibit high site fidelity. This is likely to prevent them from recolonizing habitat that become 374 375 reconnected due to dam removal and improved/mitigated river operations.

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Fig. 1 The Emory River sub-basin. The black circles signify the two tagging locations used in this study.





Fig. 2 The median (solid black line) movement (m), minimum and maximum movement (dotted black line) of PIT tagged Sickle Darters in relation to discharge (observed throughout the study.





Fig. 3 The frequency of movement upstream (positive) and downstream (negative) by PIT-tagged Sickle Darters
 throughout the study.

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**Table 1** Results of best subsets multiple linear regression modeling as a variable selection procedure for movement (response variable) by the Sickle Darter in the upper Emory River sub- basin at three temporal scales (1-day, 3-day, 7-day). The top 5 models are shown that achieved the lowest AIC<sub>e</sub>, lowest Mallow's Cp statistic, and highest adjusted  $R^2$ . Variables retained for interpretation had variance inflation factors (VIF) < 4.0. Assumptions of regression analysis were met by the top model. Discharge =median discharge, Precip = total precipitation, Daily TempChange = daily temperature change, MeanWaterTemp = mean water temperature

Variables included in Model		$\Delta AIC_{C}$	Mallows'C(p)	Adj. R <sup>2</sup>	Number of Model Parameters
1-day					
Discharge	6.14	0.00	1.92	0.39	1
Discharge, Precip	7.80	1.66	2.13	0.58	2
DailyTempChange	7.83	1.69	5.04	0.07	1
Precip	7.90	1.76	5.19	0.05	1
DailyTempChange, Discharge	8.73	2.59	2.65	0.52	2
3-day					
Discharge	4.76	0.00	0.00	0.38	1
DailyTempChange	5.95	1.20	2.59	-0.03	1
MeanWaterTemp	6.11	1.35	2.99	-0.09	1
Precip	6.16	1.40	3.13	-0.11	1
Discharge, Precip	7.00	2.24	1.35	0.37	2
7-day					
Discharge	8.39	0.00	11.88	0.69	1
Discharge, Precip	8.44	0.05	3.42	0.89	2
DailyTempChange, Discharge, Precip	9.41	1.03	3.03	0.94	3
DailyTempChange, Discharge	11.10	2.72	11.17	0.74	2
Discharge, MeanWaterTemp	11.24	2.85	11.79	0.73	2

6	1	6

across 3 temporal scales (1-day, 3-day, 7-day). Results shown are for the best model from Table 2. Root MSE= root mean square error, Stand.  $B_i$  = standardized beta coefficient, VIF = variance inflation factor. The +/- sign for t-value indicates the direction of the association between the environmental covariate and distance of stream occupied. Discharge =median discharge. Models: Temporal Scale Variable P-value Stand. Bi. VIF t-value 1-day Root MSE=2.36 Intercept 2.72 0.05 0.00 0.00 Discharge -1.82 0.14 -1.08 3.44 3-day Root MSE=7.21 Intercept 0.66 0.56 0.00 0.00 Discharge 0.16 -1.11 2.65 -1.88 7-day Root MSE=1.09 Intercept 2.01 0.140 0.00 0.00 Discharge -6.51 < 0.01 -1.44 2.51

Table 2 Analysis of variance results for best subsets MLR for movement (response variable) by the Sickle Darter

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Fig. 5 The home range estimates for the 6 PIT-tagged fish from the Rock Creek site. Each fish's home range
 displays the 90% core range and 50% core range.



**627** Fig. 6 Plot of estimated home range  $(m^2)$  and PIT-tagged fish size (mm).