FINAL PERFORMANCE REPORT

Federal Aid Grant No. F21AP03618 (E-95-R-1)

A survey for *Triaenodes tridontus* (Leptoceridae) in SE Oklahoma

Oklahoma Department of Wildlife Conservation

January 1, 2022 - December 31, 2022

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State: Oklahoma **Grant Number:** F21AP03618 (E-95-R-1)

Grant Program: Cooperative Endangered Species Conservation Fund, Traditional Conservation

Grant Title: A survey for *Triaenodes tridontus* (Leptoceridae) in SE Oklahoma

Grant Period: January 1, 2022 — December 31, 2022

Principal Investigator: Rickey D. Cothran, Department of Biological Sciences, Southwestern Oklahoma State University, Weatherford, Oklahoma

Executive Summary

We provide results for an extensive survey targeting the three-toothed long-horn caddisfly, *Triaenodes tridontus*, in eastern Oklahoma. The holotype for the species was collected in 1934 but it has not been collected since that date. We focused our efforts on southeast Oklahoma where the holotype was collected but also surveyed sites along the Spring River in northeast Oklahoma. The survey, combined with similar work conducted in the spring and summers of 2017 and 2018, represents a comprehensive attempt to collect the species covering 39 sites across three years and two seasons (spring and summer). A combination of active and passive light traps were used to collect adults and water quality data was collected at most sites to examine correlations with species occurrences. We did not collect any confirmed *T. tridontus* in the survey. We did collect two species, *T. ochraceus* and *T. aba* for which there are no records in Oklahoma but the species are found in neighboring states. Interestingly, the vast majority of individuals collected were females. This presented challenges in determining species given that many species defining characters are specific to males. Future studies that develop barcoding methods for species identification in the genus may allow researchers to circumvent such problems in the future.

Objectives:

1) We will attempt to re-collect the Three-toothed Triaenodes caddisfly (*Triaenodes tridontus*) in Pushmataha County and surrounding counties. We will focus our effort on the Kiamichi and Little River drainages.
2) We will describe habitat characteristics and other species associated with occurrences of *T. tridontus.*

Summary of Progress:

Project Background

Triaenodes tridontus (Leptoceridae) is a Species of Greatest Conservation Need in Oklahoma. This species is known from Pushmataha County where the holotype was collected in 1934 (Moulton and Stewart 1996)—but the occurrence lacks location data other than the county (checked by Vaughn 1992). A more recent survey of 10 sites on the mainstem of the Kiamichi River failed to find the species (Vaughn 1992). The IUCN lists the species as extinct and *T. tridontus* has a state rank of SH (state historic) because the only record is over 80 years old.

We surveyed sites in southeast Oklahoma for *T. tridontus* and other caddisfly and mayflies of conservation concern in the spring and summers of 2017 and 2018 (Cothran et al. 2019). We used a combination of manned (attendance at the blacklight trap) and unmanned (blacklight traps deployed and picked up hours later) to survey for adults. We were unsuccessful in collecting *T. tridontus* during those surveys. We did collect individuals of the genus and most of the collections were made during our spring field trips (May). Our approach was coarse to maximize success of collecting various target species in the 2017-2018 survey. Given our results and what little has been reported in the literature on flight patterns; we focused efforts reported here during the months of April and May (Moulton and Stewart 1996; Manuel 2010).

We surveyed for *T. tridontus* in streams located in the Ouachita Mountain-Arkansas River Valley-Gulf Coast Plain Conservation Region located in southeast Oklahoma. We collected water quality data and site information at each location to associate any found occurrences of *T. tridontus* with key ecological variables.

Methods

Surveys were conducted from April to mid-May 2022. This work complements a previous study that focused on the same region where collections were made in 2017 and 2018 from late-May through mid-August (Cothran et al. 2019). At most sites, water quality data (pH, dissolved oxygen, conductivity, total dissolved solids and temperature) were collected in triplicate the day of or the day after light trapping. Sample sites were most often bridges located on small streams in the study region.

Larval *Triaenodes* build cases out of vegetation, so we focused efforts on sites that had access to submergent or emergent vegetation (Manuel 2010). Unmanned light traps (UMLT) consisted of small plastic boxes (13 x 13 x 3 cm) fitted with a 5mm mesh lid. The metal mesh lid prevented larger insects from entering the trap to avoid collecting large numbers of non-target taxa. The trap was filled with 70% ethanol. A hand-held, battery operated blacklight in a small ziplock bag was placed on top of the trap. Traps set at dusk and were left out for ~5 hours. Manned light traps (MLT) consisted of blacklights attended by two to three researchers. We selectively targeted longhorn caddisflies (Leptoceridae) at manned light traps. In addition to 2022 collections, samples from a few light traps that were collected during the 2017-2018 survey (Cothran et al. 2019) were also processed to species as part of this project.

For identifications we first sorted caddisflies into families focusing on longhorn caddisflies. Leptocerids were then identified to genus using characteristics provided by our taxonomic expert Robert Wisseman (Senior Scientist at Aquatic Biology Associates, Inc.). This consisted of a combination tibial spur count, forewing M stem, shape of apex of forewings, mesoepimeron inspection, and katepisternum shape. Individuals identified to genus were sent to Robert Wisseman who used Manuel (2010) for species identifications. Specimens will be deposited in the Colorado State University CP Gillette Museum of Arthropod Diversity.

Results and Discussion

We collected many individuals in the genus *Triaenodes* in the 2017-2018 and 2022 surveys (Table 1). Interestingly, most of the individuals collected were females. The current taxonomy of females is based on genital character, which requires the abdomens of selected specimens to be cleared (Manuel 2010). Moreover, the genitalia of female specimens is variable and some species do not have species descriptions based on females. Collectively, this made species identification based on the specimens collected difficult. Robert Wisseman suggested DNA barcode verification for some of the specimens, which is outside the scope of the current project. Some females and all males were identified with confidence.

Females identified to the *Triaenodes (Triaenodella)* Injustus Group (Table 1), which is near *T. tridontus* were significantly different than *T. tridonta* based on Manuel (2010). However, there is no comparative material to work with and the range of variation in genital morphology of this group is unknown. The two females were collected using an UMLT at the Cloudy Creek site (Table 1) on 16 May 2018. If possible, Robert Wisseman suggested DNA barcoding for these specimens.

Individuals identified to the *Triaenodes (Triaenodella)* Perna Group could not be identified to species because not all females for this group are known (Table 1).

The two females of *Triaenodes (Triaenodes) aba* collected at the Kiamichi River, Fields Road site (Table 1) are unknown to Oklahoma according to Manuel (2010). There are records from Arkansas. Based on this information, our collections suggest a range extension in the southern part of the species' range.

Similarly, our collections of *Triaenodes ochraceus* at the Rock Creek site (Table 1) are apparently the first records for Oklahoma, although the species is known from Texas and Arkansas.

Acknowledgements

I thank Dr. Peter Grant and Casady Sublett for assistance in the field. Many thanks to taxonomic expert, Robert Wisseman at Aquatic Biology Associates, Inc. for providing species identifications of collected *Triaenodes*.

Recommendations:

A peculiar outcome was the asymmetry of collected individuals between the sexes. Out of the 123 individuals collected in the genus *Triaenodes* only 12% were males. The vast majority of the specimens were collected UMLTs but even at the MLTs only 18% of individuals collected were males. It is unclear whether this pattern is due to differences in how attracted the sexes are to the blacklight or reflects extreme skewness in adult sex ratios. Importantly, this limits outcomes based on surveys that use blacklights because many of the species identifying characters are known from males.

If properly developed, DNA barcoding technology may overcome problems with sex-specific collection. However, a literature search uncovered no genetic markers that could be used to identify *Triaenodes tridontus* to species. Investment in such markers would greatly improve our ability to monitor for this species.

Significant Deviations:

None

Equipment Purchased (Cumulative):

No equipment exceeding $5,000 was purchased under this grant.

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References:

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Manuel, K. L. 2010. The longhorn caddisfly genus *Triaenodes* (Trichoptera: Leptoceridae) in North America. The Caddis Press, Columbus, OH.

Moulton, S. R. and K. W. Stewart. 1996. Caddisflies (Trichoptera) of the interior highlands of North America. American Entomological Institute.

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Table 1 (Excel File). Summary of *Triaenodes* collections in eastern Oklahoma for 2017, 2018 and 2022. For many of the 2022 collections, water quality results, averages from three replicates, are provided. For 2022 collections, dates of manned (MLT) and dates and number of unmanned light traps (UMLT) are provided. For 2017 and 2018 results are for light traps that were not completed to report in Cothran et al. 2019. Numbers in taxonomic cells represent the number of a particular sex collected for the group. Shaded cells are outcomes for MLTs. No *Triaenodes tridontus* were collected in the surveys.

Table 1. Summary of Triaenodes collections in eastern Oklahoma for 2017, 2018 and 2022. For many of the 2022 collections, water quality results, averages from three replicates, are provided. For 2022 collections, dates of manned (MLT) and dates and number of unmanned light traps (UMLT) are provided. For 2017 and 2018 results are for light traps that were not completed to report in Cothran et al. 2019. Numbers in taxonomic cells represent the number of a particular sex collected for the group. Shaded cells are outcomes for MLTs. No Triaenodes tridontus were collected in the surveys.

										Light Traps 2022		
	Location		2022 Water Quality Data								UMLT	
Site	Latitude	Longitude	Date	Time start	pH	Dissolved Oxygen (mg/L)	Conductivity (uS)	Total Dissolved Solids (mg/L)	Temperature (°C)	MLT Date	Date	Number
Dry Creek	34.62708	-95.24125										
Kiamichi River at Big Cedar	34.63876	-94.65446										
Peal Creek	34.56705	-95.33856										
Pine Creek	34.4089	-95.60351	4-2-22	10:06	5.84	11.10	53.33	27.00	14.67	4-15-22		
Kiamichi River at Fields Road	34.63665	-94.61503	5-11-22	13:40	4.98	10.48	17.00	9.00	21.32	4-23-22		
Middle One Creek	34.36494	-95.37117	4-16-22	9:30	5.22	6.92	39.00	20.00	17.21			
Crum Creek	34.52734	-95.47968	4-2-22	12:15	5.50	11.04	25.67	13.00	14.56	4-1-22	4-15-22	2
Little River at Ludlow	34.5395	-94.84793	4-2-22	19:39	5.17	10.09	19.00	10.00	15.91	4-2-22		
Little River at Nolia	34.49678	-95.12699	4-2-22	17:48	5.45	11.17	22.00	11.00	15.11		4-2-22	2
Cloudy Creek	34.3246	-95.22344	4-16-22	11:40	5.38	6.54	53.00	27.00	19.14	4-22-22		
Holly Creek	34.3294	-95.1502	4-16-22	13:16	5.31	7.97	39.67	20.00	17.52			
Yashau Creek	34.10137	-94.76648	4-16-22	16:54	6.33	8.11	111.00	55.00	17.67		4-16-22	1
Lukfata Creek	34.11992	-94.81485	4-16-22	17:53	6.50	8.69	153.67	76.67	17.37	4-16-22		
Briar Creek	34.10647	-94.78402										
Glover River	34.09771	-94.90253									4-16-22	2
Mountain Fork River at State Line Road	34.64193	-94.45735	4-23-22	18:37	4.75	9.79	20.00	10.00	18.30			
Spring Creek 1	36.17616	-94.86251										
Spring Creek 3	36.0906	-95.015										
Spring Creek 4	36.10405	-95.0961										
Spring Creek 5	36.12624	-95.20821										
Gaines Creek	34.81029	-95.34853	5-10-22	16:20	5.62	8.39	59.00	29.67	25.51		5-10-22	2
Buffalo Creek	34.79615	-95.48315	5-10-22		6.04	6.63	64.67	32.67	23.57	5-10-22		
Small Pine Creek	34.80821	-95.34591	5-10-22	15:30	5.56	9.46	42.33	21.00	25.59	None	None	
Jackson Creek	34.73117	-95.09524	5-11-22	10:20	6.09	9.01	55.33	29.33	21.01		5-10-22	1
Little Buffalo Creek	34.72765	-95.18259	5-11-22	11:05	5.57	8.58	57.00	28.00	20.72		5-10-22	1
Rock Creek	34.76039	-95.08394	5-11-22	9:40	5.26	9.52	26.00	13.00	17.07		5-10-22	2
Bohanon Creek	34.70187	-94.91602	5-11-22	12:13	5.23	8.43	36.33	18.00	24.03	None	None	
Push Creek 1	33.73849	-94.69431	5-12-22	10:48	6.12	2.06	89.33	45.00	26.08	none	none	
Push Creek 2	33.71318	-94.60739	5-12-22	14:42	5.86	5.83	159.00	80.00	24.69	none	none	
McKinney Creek	33.74945	-94.52131	5-12-22	14:48	5.21	3.83	153.00	76.33	23.42	none	none	
Walnut Bayou	33.69784	-94.52097	5-12-22	15:32	7.05	7.43	99.00	49.00	30.28	none	none	
Ward Creek	34.39959	-94.6936	5-13-22	11:58	5.25	8.64	26.00	13.00	19.04		5-11-22	1
Middle Carter Creek	34.26138	-94.79376	5-13-22	9:57	5.03	7.75	36.00	18.00	22.15		5-11-22	1
Eagle Creek	34.54818	-94.69202	5-13-22	12:55	5.24	9.33	17.00	8.33	20.92	5-11-22		
Little Pine Creek	34.33013	-94.8241									5-11-22	1
North Carter Creek	34.28723	-94.82594									5-11-22	1
Cedar Creek	34.18245	-94.8517									5-12-22	1
Tidwell Creek	34.1683	-94.86768									5-12-22	1
Canyon Creek	34.30904	-94.81319									5-11-22	1
Caney Creek	34.15783	-94.88307									5-12-22	1

	T. injustus						*T. tardus*						*T. (Trianenodella)* most likely *tardus*						*T. ignitus*					
	2017		2018		2022		2017		2018		2022		2017		2018		2022		2017		2018		2022	
Site	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female
Dry Creek				3				2								1								
Kiamichi River at Big Cedar																								
Peal Creek																								
Pine Creek										1														
Kiamichi River at Fields Road										3						3								
Middle One Creek			1				3			3											1	1		
Crum Creek																								
Little River at Ludlow																								
Little River at Nolia	1	2		10				2																
Cloudy Creek				18						9		2				1								
Holly Creek										5														
Yashau Creek																								
Lukfata Creek																								
Briar Creek																1					1			
Glover River																								
Mountain Fork River at State Line Road										4														
Spring Creek 1																1					1			
Spring Creek 3										1						1								
Spring Creek 4										2						11								
Spring Creek 5																6								
Gaines Creek												4												
Buffalo Creek												3						1						
Small Pine Creek																								
Jackson Creek						1						1												
Little Buffalo Creek																		6						
Rock Creek																		1						
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North Carter Creek												8						3						
Cedar Creek																					1			
Tidwell Creek																								
Canyon Creek																								
Caney Creek																								

	T. ochraceus						*T. cumberlandensis*						*T. aba*						*T. (Triaenodella)* Injustice Group near tridonta						*T. (Triaenodella)* Perna Group					
	2017		2018		2022		2017		2018		2022		2017		2018		2022		2017		2018		2022		2017		2018		2022	
Site	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female
Dry Creek																														
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Crum Creek																														
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