

Springflow Habitat Protection Work Group

June 3, 2020 2:00-4:00pm

Today's Meeting

- Clarify and refine
 - The Implementing Committee should ensure a technical evaluation is undertaken of potential <u>impacts of predicted</u> <u>extended periods of flow below 80 cfs on Comal Springs riffle</u> <u>beetle populations;</u>

Agenda Overview

- Confirm attendance
- Meeting logistics
- Public comment
- Presentation and discussion
 - Texas Parks and Wildlife 2011 and 2014 Comal Springs mapping and how that relates to occupied Comal Springs riffle beetle (CSRB) habitat
 - Preliminary results of CSRB occupancy study
 - How recent drought (2011-2014) has impacted CSRB populations
- Public comment
- Future meetings

Confirm attendance

Meeting logistics

- Virtual meeting logistics
 - Mute
 - Raise Hand
 - Chat / Asking questions
 - Meeting recording



• Meeting points of contact

- Meeting access
 - Victor Hutchison (vhutchison@..)
- Technical questions
 - Victor Hutchison (vhutchison@..)
 - Martin Hernandez (mhernandez@..)
- Participant monitor
 - Kristy Kollaus (kkollaus@...)
- Chat and Q&A monitors
 - Kristina Tolman (ktolman@...)
 - Damon Childs (dchilds@...)

Public comment

Comal Springs Mapping as it Relates to Comal Springs Riffle Beetle Habitat





Chad Norris Groundwater Resources Coordinator Water Resources Branch Texas Parks and Wildlife Department <u>Chad.Norris@tpwd.texas.gov</u> 512-847-5078

Springflow Habitat Protection Workgroup Microsoft Teams My House June 3, 2020

Comal Springs Mapping

- Mapping performed in 2012 at 240 cfs
- 425 springs (orifices, lines, and polygons) mapped
- Location data included Trimble GPS and station on measuring tape
- Water quality, elevation, and substrate data
- Elevations based on EAA benchmark monument system in Landa Park and water surface elevation
- Flow-partitioning data gathered by EAA staff
- Attempted repeat during drought in 2014

April 2012 – 240 cfs

- 425 Springs Features Total
 - Points (Green)
 - Lines (Purple)
 - Polygons (Orange)
- Spring Runs
 - Total 113 (27%)
- Spring Run 1-21
- Spring Run 2 14
- Spring Run 3 57
- Spring Run 4 6
- Upper Spring Run 13
- W Shore 142 (33%)
- Landa Lake 62 (15%)
- Spring Island 101 (24%)

- What Springs will remain flowing?
- Do these springs contain populations of CSRB?
- What is habitat like at these springs?

Comal Springs Riffle Beetle

- Habitat closely associated with spring openings
- Survived Drought of Record mechanism unknown
 - Signs of genetic bottleneck
- Biomonitoring at "Representative Reaches" Spring Run 3, Western Shoreline, and Spring Island
- No thorough sampling has been performed to define range in system
- Early analysis of CSRB habitat during EARIP focused on protecting Spring Run habitat – "conservative approach"

Comal Springs Riffle Beetle Assumptions

- Later analysis assumed Western Shoreline and Landa Lake habitats would remain at 30 cfs and sustain CSRB through proposed flow regime
- CSRB habitat evaluations in Hardy (2009) assumed that areas G through L (i.e. Western Shoreline, Lower Landa Lake, and Spring Runs) contribute 90 percent of the total river discharge at flows less than 225 cfs.
- Hardy (2010) "Springs along the western margin of Landa Lake are anticipated to provide adequate habitat during the lower flow regime and in our opinion as flow increases to the 80 cfs range that the lower extent of Spring Runs 1, 2 and 3 will be hydraulically connected to Landa Lake given expected lake elevations and lake bathymetry."

*Hardy, T.B., 2009, Technical assessment in support of the Edwards Aquifer Science Committee "j" charge—Flow regime evaluation for the Comal and San Marcos river systems: Prepared for the Edwards Aquifer Recovery and Implementation Program, River Systems Institute, Texas State University.

*Hardy, T.B., K. Kollaus, and K. Tower. 2010. Evaluation of the proposed Edwards Aquifer recovery implementation program drought of record minimum flow regimes in the Comal and San Marcos River Systems. December 28, 2010. http://earip.org/Hardy/EARIP_1-6-2010_Draft_03.pdf

Comal Springs Hydrodynamics

Figure 17. Spatial location of spring inflow nodes within Landa Lake of the Comal River system used in the hydrodynamic modeling.

*Hardy, T.B., 2009, Technical assessment in support of the Edwards Aquifer Science Committee "j" charge—Flow regime evaluation for the Comal and San Marcos river systems: Prepared for the Edwards Aquifer Recovery and Implementation Program, River Systems Institute, Texas State University.

Comal Springs and Landa Lake, Comal County, Texas

Flow-Partitioning

- Aug 2013 Sept 2014
- 140 68 cfs
- **Spring Island Area** provides 40-50% of total flow
- Landa Lake % increases as total flow decreases

Motor Map of Comal Springs and the surrounding area USGS Gauge including tributaries, stream runs and USGS gauges. 150 300 450 600 Datum: NAD 1983 UTM Zone 14N. Map scale 1:7,500

*Rohan, Catherine, 2014. Analysis of flow at Comal Springs, Comal County, Texas. University of Texas at Austin.

% Contribution of Springflow at flows below 140 cfs

TEXAS

PARKS &

WILDLIFE

120

- GW flows from Artesian
 Block to Comal Springs
 Block at normal and
 low flows
- < 100 cfs, water in
 Comal Springs Fault
 block bypasses Comal
 and travels to San
 Marcos Springs
- Artesian Block feeds
 Landa Lake springs
- W Shore Artesian, Water Table, or transition zone?

Spring Run 3 ceases - 620'

Flow at USGS gage ceases - 619'

Historic low 613.34' on 8/21/56

Limited data at low end

Data from USGS and TWDB

*LBG Guyton and Associates. 2004. Evaluation of augmentation methodologies in support of in-situ refugia at Comal and San Marcos Springs, Tx, prepared for the Edwards Aquifer Authority. 192 p.

Spring features ≤ 620'

195 features

- W Shore 7
- Spring Run 3 6
- Spring Island 95 (49%)
- Landa Lake 62
 (32%)

Spring features ≤ 619'

152 features

- W Shore 3
- Spring Run 3 2
- Spring Island 71 (47%)
- Landa Lake 61 (40%)

April 2012 - 240 cfs

- 425 Springs Features Total
 - Points (Green)
 - Lines (Purple)
 - Polygons (Orange)
- Spring Runs
 - Total 113 (27%)
- Spring Run 1- 21
- Spring Run 2 14
- Spring Run 3 57
- Spring Run 4 6
- Upper Spring Run 13
- W Shore 142 (33%)
- Landa Lake 62 (15%)
- Spring Island 101 (24%)

Sept 2014 – 80-90 cfs

- 97 Springs Total (41% of '12)
- West Shore
 - 54 (38% of '12)
 - 45 described as seeps
- Spring Run 1 - 10 (47% of '12)
- Spring Run 2 - 4 (28% of '12)
- Spring Run 3 - 29 (51% of '12)

*Rain ended effort early

Western Shore Spring

Comal Springs Riffle Beetle

Effects of low flows on Comal Springs Riffle Beetle

- What Springs will remain flowing? Landa Lake and Spring Island, maybe Western Shoreline? Is elevation data helpful? Geophysical data needed?
- Do these springs contain populations of CSRB? Hard to say, more sampling needed
- What is habitat like at these springs? Is the habitat conducive to CSRB? Geophysical data?

Comal Springs Mapping as it Relates to Comal Springs Riffle Beetle Habitat

Chad Norris Groundwater Resources Coordinator Water Resources Branch Texas Parks and Wildlife Department <u>Chad.Norris@tpwd.texas.gov</u> 512-847-5078

> Microsoft Teams My House June 3, 2020

Occupancy and Abundance of the Comal Springs Riffle Beetle (Heterelmis comalensis)

> Kayla Robichaux and Dr. Weston Nowlin

CSRB Populations

- Found primarily at Comal Springs
- Use lures (poly-cotton cloths) to monitor and collect CSRB
 - Useful technique
- CSRB population estimates and site occupancy - limited quantitative examination
- Estimate occupancy and population size of the CSRB at Comal Springs sites using N-mixture models

Occupancy Modeling

- Often cannot exhaustively survey an area
 - "Shy" organisms and/or low population densities
 - CSRB like this?
- Occupancy models
 - Accounts for imperfect detection
 - "Detection error"
 - Determine the probability of presence or abundance at a site
 - Replication over space and time
 - Extension of GLMEMs
 - Use detect/non-detect data
 - Utilize environmental covariates influence occupancy

N-Mixture Modeling

- Used in conjunction with occupancy estimates
 - Estimate abundance from count data (imperfect detection)
 - Replicated over space and time
- Probability of detection and count data at sites used in model
 - Two linked GLMs
- Open and closed populations
- Gives you the abundance or population size at each site and potentially across sites
 - Only where you sampled
- Can also include covariates

Our Study

Goals

- Estimate the occupancy and abundance of CSRB across Comal Springs
- Identify significant covariates that aid in prediction of occupancy and population size across Comal Springs

Sampling Design

- Stratified randomized design
 - Spring openings/discharge points using standard lures
 - Hydrological "units"
 - Spring Runs 1, 2, 3, 4
 - Western Shoreline
 - Spring Island
 - Landa Lake
- Mapped >500 spring openings (2018)
- Randomly selected sites, >3 m apart (Huston et al. 2015)
- n = 85 sites
 - Sites per area based on # of springs in area (5 to 33)
- Avoided biomonitoring sites

$$\frac{n = 85 \text{ Springs}}{\text{SR1-3} = 23 (27\%)}$$

WS = 33 (39%)
SI = 12 (14%)
LL = 12 (14%)
SR4 = 5 (6%)

FID	Location	SID	Picture	GPS#	map feature Description of Lure Placement
	0,000 42				
1	SR1	6	yes		poly upwelling, in front of small boulder between 2 huge boulders, triangulate: 2.2 m from tag 12, 2.5m from tag 9, 2.5 ft to the right of giant rock when looking from bank
2	SR1	9	no		point upwelling, 1.7 ft away from wall tag
3	SR1	21	yes		point FL, 2 ft to the left of corner of wall (deep in wall), fern covering
4	SR1	45	yes		point FL, in crevice of fallen tree about an arms length back
5	SR1	48	yes		point FL, under tag, to the left of fallen tree, to the left of fern, lure a forearm in
6	SR1	56	no		point FL, next to big tree, tag on tree, to the left of gauge, forearm length to left of "rock ledge"
7	SR2	68	no		point RS of SR2, directly under tag, near surface, wedged at an angle, (water level lower now, had to place in slightly different spot)
8	SR2	75	yes		point FL, no tag, at base of anacua tree 1 ft to left of large root in small opening
9	SR2	81	yes		point FL, LS, directly under tag straight back about a forearm length in (watch out for poison ivy)
10	SR2	93	yes		point RS, FL, directly under tag behind hanging roots, near white boulder, placed on upwelling half a forearm length in
11	SR2	99	yes		point tag on rock further out in channel, almost exactly between tag 96 and 105, large flow, in bank 4" in under hanging roots under rock pile. Cove in bank on right side of SR2
12	SR3	117	yes		point FL, at headwaters of SR3 in right corner, to the right of the edge of wall
13	SR3	135	yes		line about 1 ft to left of white cyllinder, big white rock on top
14	SR3	138	yes		point FL, below, left of tag, in recess under rock, half a forearm in
15	SR3	153	yes (2)		poly sand upwelling in center of channel to the right of tag 151, straight out from white pipe with two red boulders on top
16	SR3	156	yes (2)		point further to the right than previously placed, in upwelling w/ 2 big boulders on sides
17	SR3	163	no		point under tag to left of large stump, sandy upwelling with red stone on left
18	SR3	174	yes		point under footbridge, under tag to the left behind white rock
19	SR3	190	yes		point FL, lure beneath tag located on roots, seep
20	SR3	195	yes		line FL, lure to the left of tag under tree, about a forearm back
21	SR3	198	yes		line FL, lure to the left of tag about 1.5 ft under roots, shallow area wedged into rock, white rock in front
22	SR3	240	yes		line upstream of bridge, no tag, found to the left of Turks cap in rooty area, uppercut to the right of boulder
23	SR3	246	yes		point downstream of bridge, to the right of Turks cap, right edge of tree cage on bank, straight down in crevice
24	ws	271	yes		line After 276, No tag, on the left side of large boulder w/ tree stump above it, near Ligustrum (?) and bank, pink rock lost in crevice
			100.0000		
25	ws	276	yes		point FL, Before 2/1, under tag W/ white rock on top, not very far in. To the right of Turks cap, next to big log sticking out into middle of channel down almost in SR3
26	WS	285	(2)		line FL, to the left of tag 285 at the point in the alcove under Anacua tree about an arms length in
27	ws	288	yes (2)		point FL, tag on Anacua tree, to lett of tag about 3tt
20		201	(2)		FL, tag on rock, spring below tag to the right of rock (1 rt right of tag), deep spring, wedged fure between rock and against submerged roots, almost an arm length deep, no pir
28	WS	291	yes (2)		
29	WS	297	yes		point FL, to right of 291 under trees, left side of orfifce under rock
30	WS	318			point pr. to the right of took shell winder tag, about 1/2 forearm in
51	ws	522	yes		point rt, tag moden by trees between 321 & 324 tags, minder tag nanging routs, nan arms lenth in tag underspatial Angeue tags to the two initials of 250 on tag of section ground during larged an left side of tag in root wad. 90 degree and down from tag tags tagt tagk 5 inches
22	MC	261			lag underneaut Anadua itee, to the right of 555 on top of rock rating upward, the plated on left side of tag in root wad, 50 degree angle down from tag to fed tack 5 inches,
52	vvs	201	yes		point rout daw to reit o indices
20	MIC	272			to the right of 0505 big pole, tag of rock, spring to the right of tag under Analua deep in track in rock, 5 it down between 2 boulders (need someone to hold legs so don't hold
24	VV5	202	yes		point lawdy)
25	VVS WS	202	yes		point tag on sump in elephant ear, under private property sign on rence, fue to the fert of tag, remove an rubber to get to base on on ent
- 35	VV3	307	yes		point to the right of sos in elephant ear, tag on nor stump, use to the rest side of stump in noots wedged up against elephant ear stark
26	MIC	202			to the right of elephant ear stand in roots @ large tree not far from 587, tag down on roots hear water to the left of trunk, fure 2 it to right of tag on left side of noie, no pink
27	WS	393	yes		mile jook
3/	WS	41/	yes		June EL just to the right of feare, the on tree. 2 ff to the left of the log is not a bout a
20	VVS	432			line 1.5 ft to left frag on tree, 2 into the left of tag in foots about a fiditio didefineatin
39	VVS	444	yes		mine 1.3 in to lead of the final to red mining to the left of tag 1 ft lung in the expering and back to the right towards the tag
40	VVS	440	Nor		point [La colloff ide of reach under more function costs.
41	VVS	4/9	yes		Jung jung a bout a
42	VVS M/C	405	yes		nice just under tag, adout a flatiu in, tag covereu in algae on other side of fock nonf 462
1 73	vv3	405	yes		pointline under rug on rice.

- Lure deployed at each point for a 5-week period (Huston et al. 2018)
 - 4 sampling events
- CSRB adults and larvae enumerated and gently put back
 - Wait 1 week, redeploy
- Environmental covariates
 - pH, SpCond, Temp, DO
 - Length of deployment
 - Substrate composition (gravel, sand, silt, etc)
 - Pres of roots/terrestrial OM
 - Water velocity
 - Pres of other inverts (*M.p.* and *S.p.*)
 - Water depth
 - Elevation
 - % biofilm cover on lure (0-4)
 - Precipitation
 - Comal discharge

Status and Preliminary Results

- Field collections complete (Nov 2019)
- Data analysis stage
 - GLMs used to assess relationships between CSRB adults/larvae and environmental predictors
 - Reduce predictors/covariates to include in final occupancy and *N*-mixture models
 - Pearson correlations among environmental predictors
 - ANOVAs for differences among site covariates/predictors

Status and Preliminary Results

Significant Environmental Predictors

<u>Adults</u>

- Spring elevation (+)
- Water depth (-)
- DO (-)
- Presence of roots (+)

<u>Larvae</u>

- Water depth (-)
- Presence of roots (+)
- Percent coverage of biofilm (+)

Key Findings (So Far)

• Spatial variation in abundance (and occupancy)

- Higher elevation, riparian connection
- Upper springs and WS
- Difference in adults and larvae
- •CSRB adults and larvae at SR4
- •Complete occupancy and *N*-mixture models

How has recent drought impacted CSRB populations?

Will Coleman

PhD Candidate, Texas State University

Co-Advisors: Dr. Chris Nice and Dr. Benjamin Schwartz

Ecological and Evolutionary Genomics of Groundwater Biodiversity

- My dissertation research is focused on population genetics
 - Heterelmis beetles across the southwestern U.S.A. and Mexico (see map)
 - *Lirceolus* isopods, including the Texas Troglobitic Water Slater (*L. smithii*)
 - Comal Springs Dryopid Beetle (*Stygoparnus comalensis*)
- Is nominal taxonomy supported by molecular data?
- What are species ranges? Where are the boundaries separating populations?
- Comparative approach within and among taxa

Collection sites for *Heterelmis* genus-level project

What can we learn from genetic data?

- Diversity of populations
 - Genetic variability within and among populations
 - <u>Heterozygosity</u>: two different alleles at a locus (Aa)
- Is there population structure?
 - Presence or absence of gene flow
- How many beetles are there?
- <u>Effective population size (Ne)</u>: the effective number of breeding adults in a population

Heterelmis beetles are <u>diploid</u> organisms. Individuals have two alleles.

Past genetic studies of CSRB

- 2008 T. Gonzales M.S. Thesis (unpublished data)
 - mtDNA study (one marker at a single locus)
 - Modest amount of genetic variation among Western Shore, Spring Island and San Marcos Springs populations
 - Populations from spring Runs 1, 2, and 3 were genetically invariant
- 2016 Lucas et al. (Freshwater Biology)
 - Next-generation sequencing analysis of the same individuals (545 markers)
 - Little evidence of subpopulation structure, 'pervasive gene flow'
- But what about Ne?

For my genus-level analysis, I have obtained genotype data for ~15,000 markers

Estimating Effective Population Size (Ne)

- Estimating Ne from a single sampling period is weak
- A temporal sampling approach is a much more effective way of obtaining estimates of Ne
 - Estimate genetic drift in the generations between sampling events
 - This method is robust because
- Variance in allele frequency is a function of population size ...

Genetic Drift and Population Size

- <u>Genetic drift</u>: random change in allele frequencies in a population
- The variance in allele frequencies over generations is a function of population size
 - Requires more than one sampling period!
- Let's do some simulations

Variance in allele freq. over time: N=10, 1 rep

Variance in allele freq. over time: N=10, 10 reps

Variance in allele freq. over time N=100, 10 reps

Variance in allele freq. over time N = 1000, 10 reps

What does it mean?

- Variance in allele frequency is a function of population size
- Genetic drift has a larger impact on smaller populations
- With a temporal sampling approach, we can obtain allele frequency estimates at multiple times and use these to calculate effective population size
- I will do this using...

Approximate Bayesian Computation

- Simulate a population using a model
- Do this about one million times considering a range of possible Ne values
- Ne=10, Ne=11, Ne=12... Ne=1,000,000
- Calculate summary statistics for each of these simulations
- Examine where the <u>observed summary statistics</u> of the fall within the distribution of <u>simulated summary statistics</u> and <u>possible Ne values</u>

Summary Statistics

```
H - average Heterozygosity
p - average minor allele
frequency
F<sub>IS</sub> - Inbreeding coefficient
F<sub>ST</sub> - differentiation between
time 0 and time t
```

Get Data Summary Statistics H, p, F_{IS}, F_{ST} etc.

Draw parameter values randomly from a uniform distribution Simulate same markers as real data Simulate a bunch $\sim 10^6$ +

106

10

Ne

Simulate Data Under Model

Get Data Summary Statistics H, p, F_{IS}, F_{ST} etc.

Calculate Summary Statistics H, p, F_{IS} , F_{ST} etc.

Simulate Data Under Model

Get Data Summary Statistics H, p, F_{IS}, F_{ST} etc.

Calculate Summary Statistics H, p, F_{IS} , F_{ST} etc.

Examine Joint Distribution

Parameters (Ne)

Summary Stats

Simulate Data Under Model

Get Data Summary Statistics H, p, F_{IS}, F_{ST} etc.

Calculate Summary Statistics H, p, F_{IS} , F_{ST} etc.

Examine Joint Distribution

Example inference of Ne and 95% credible interval: 487 (420-572)

Approximate Bayesian Computation

- Bottom line: more simulations than you can shake a stick at
- This temporal sampling approach is robust, and it gets even better with more sampling periods (I have 3!)
- I will also implement a few methods of estimating Ne
 - Jorde and Ryman (2007) Unbiased estimator for genetic drift
 - Produces a mean Ne for your sampling period
 - Linkage Disequilibrium-based estimators (Waples 2008)
 - Watterson's theta ($\theta = 4Ne\mu$) (Watterson 1975)
- And, with my genus-level analysis of *Heterelmis*, I will be able to perform comparative analyses with closely related taxa.

Project status

Collections

Site	2007	2016	2020
Spring Run 1	20	34	34
Spring Run 2	21	34	34
Spring Run 3	21	34	34
Spring Island	21	34	34
Western Shore	29	34	34
Hotel Springs, Spring Lake	28	34	in progress

- Variance in allele frequency from...
 - 2007 to 2016
 - 2016 to 2020
 - 2007 to 2020

Preliminary analyses suggest that I will obtain sequencing data for over 15,000 loci for this project

Significance

- Estimating Ne is vital to the conservation and management of endangered species
- Temporal sampling is a robust approach, as is ABC
- Well-suited for inconspicuous organisms
- How are karst spring-adapted invertebrates affected by extreme climatic events?

Stay tuned! Results coming soon...

- Future monitoring??
- My data will answer how CSRB populations were impacted by the 2010-2015, but what about long term population trends?
- These methods would be well-suited for regularly assessing effective population size of CSRB
- Cost for DNA extraction and sequencing is \$2000/sampling period
 - That's a lot of data for your dollar

Acknowledgements

- Funding sources for this project:
 - USFWS
 - Southwestern Association of Naturalists
 - National Cave and Karst Research Institute
- Thanks to help from
 - Randy Gibson
 - Tina Gonzales
 - Chad Norris

Southwestern Association of Naturalists

Questions?

References

- Gonzales, T.K. (2008). Conservation Genetics of the Comal Springs Riffle Beetle (Heterelmis comalensis) Populations in Central Texas, with Examination of Molecular and Morphological Variation in Heterelmis Sp. Throughout Texas. MS Thesis. Texas State University, 2008.
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- Waples, R.S. and Do, C. (2008). LDNE: a program for estimating effective population size from data on linkage disequilibrium. Molecular Ecology (8): 753-756.
- Watterson, G.A. (1975). On the number of segregating sites in genetical models without recombination. Theoretical Population Biology(7)2: 256-276.

Public comment

Future meetings

- Meeting 5 TBD
 - Approve Meeting 2 Minutes
 - TBD
- Meeting 6 TBD

Thank you! eahcp@edwardsaquifer.org