1. **Title:** Using Upper Thermal Limits for Select Mussel Species to Guide Water Management Practices in the Edwards Plateau

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4. **Eligibility:** Both the Mills and USGS programs.

5. **Funds:**
Funds will go toward supporting ongoing research to study the thermal tolerance of Texas freshwater mussels. A previous study investigated the effects of temperature on the glochidial (larval) life stage of select freshwater mussels (Khan et al. 2019). Standard acute laboratory tests were run to determine the median lethal temperatures (LT50) and the lethal temperatures for 5% of the population (LT05) for various species across four basins. In my thesis project, I plan to test the thermal tolerance limits of two state-threatened species (*Lampsilis bracteata* (Texas Fatmucket) and *Cyclonaias petrina* (Texas Pimpleback)) within the San Saba River, which are presently being considered for listing under the U.S. Endangered Species Act (ESA, USFWS 2011). Lethal and sublethal responses of the glochidial and juvenile life stages will be determined using standard median lethal temperature techniques. In addition, the impacts of in situ exposures of juvenile mussels on growth rate and survival within perennial and intermittent reaches of the San Saba will be quantified and analyzed. These results will be used to determine the thermal optimums of each species, which will then be overlaid on water temperature data from the San Saba to determine if intermittency is contributing to their decline in this system and to aide in the development of eflow thresholds to protect water quantity for humans and wildlife. Such information is critical to river authorities, municipalities, and other water users likely to be impacted by ESA listings.

6. **Abstract:**
Freshwater systems are globally changing, and this can broadly be attributed to anthropogenic impacts and climate change. These issues are exacerbated within the Southwestern United States where an increasing human demand for freshwater resources coupled with climate change is resulting in more frequent periods of extreme low flows, characterized by elevated water temperatures, which can be catastrophic to aquatic biota and ecosystem function. The San Saba River, located in central Texas and part of the Colorado River basin, exemplifies this issue as recent research has shown that excessive groundwater extraction along with unique bedform geology and climate change may be resulting in as much as two-thirds of the river becoming intermittent. This change poses a threat to agricultural, residential, and wildlife needs. The latter has become an issue in the San Saba/Colorado River as it currently harbors 4 endemic mussel species that are currently under review for protection under the U.S. Endangered Species Act (USFWS 2011). There is reason to believe that elevated water temperatures associated with these intermittent periods are now impacting these species based on recently conducted thermal tolerance studies (Khan et al. 2019). However, more comprehensive testing along with in situ field monitoring is needed to determine if this is the case. To address these issues, I plan to test the upper lethal thermal tolerance limits of two species (*Lampsilis bracteata* and *Cyclonaias petrina*) within the San Saba River using standard laboratory techniques. I will measure the thermal tolerance effects of both glochidia and juveniles for each selected species across a range of common and extreme water temperatures. In addition, I plan to expose laboratory raised juveniles to perennial and intermittent regions of the San Saba to quantify and analyze growth and survival. These results will be used to determine the thermal optimums of each species, which can then be overlaid on water temperature data from the San Saba to determine if intermittency is contributing to their decline in this system. If this proves to be the case, then this data will be helpful for informing water management practices that provide for humans while ensuring the long-term persistence of rare and common mussel species in Central Texas river systems.

7. Proposed Research

a. Statement of Critical Regional or State Water Problem

Within coming years, Texas is projected to experience prolonged droughts and beyond normal temperatures due to climate change (Shafer et al. 2014). In addition, the population of Texas is exponentially growing and expected to increase 73.3% within the next 50 years (Texas Water Development Board, 2019). With this rapidly growing population comes an increased demand for water which is likely to further exacerbate anthropogenic impacts to freshwater resources that threaten both aquatic fauna and water security for the people of Texas. The San Saba River, located in central Texas and part of the Colorado River basin, exemplifies this issue as recent research has shown that excessive groundwater extraction along with unique bedform geology and climate change may be resulting in as much as two-thirds of the river becoming intermittent. Because of this, the San Saba is considered one of the most endangered rivers in the United States (Edwards et al. 1989). Intermittency and elevated water temperatures in the San Saba River pose a threat to agricultural, residential, and wildlife needs. The latter has become a high priority water management issue in this river (Randklev et al. 2018) as 4 endemic mussel species are now under review for protection under the ESA (USFWS 2011). There is reason to believe that persistent low flows and elevated water temperatures are now impacting these mussel species based on recent thermal tolerance studies (Khan et al. 2019).

Mussels are predisposed to the detrimental effects of low flow and increased water temperature due to their biology and life history. First, mussels are ectotherms which means survival, growth, and reproduction are regulated in part by external water temperature. Second, mussels are sedentary which means they are unable to move out of harm's way, especially during acute, wide-scale impacts such as stream dewatering. Finally, mussels are obligate parasites, reliant on specific host-fish for dispersal who are also adversely impacted by stream dewatering and often equally sensitive to elevated water temperatures (Gates et al. 2015). As a result of these constraints, elevated water temperatures can quickly reach uninhabitable levels during periods of low flow and depending on the frequency and magnitude can have a profound negative impact on population persistence (Khan et al. 2019).

Despite the fact that freshwater mussels are sensitive to low flow and increased water temperature, they are rarely considered during the development of environmental flow recommendations. The reason for this has to do with the fact that species-specific information on how they respond to these impacts is often unavailable. In cases where such data is available, mussels have proved critical to identifying flow thresholds across a range of discharges that are protective of both human and wildlife needs. For example, Castelli et al. (2012) modeled flow needs for the endangered Dwarf Wedgemussel (*Alasmidonta heterodon*) in the Delaware River, and was able to determine limiting temperatures that could create unfavorable conditions by using a combination of thermal tolerance and water temperature data. The researchers of this study were then able to relate this information back to flows to provide guidance on eflow needs for this species. In Texas, a process was set forth by Senate Bill 3 in 2007 for the development of flow standards that support a sound ecological environment by addressing water quantity and, to a lesser extent, quality issues. This process is also informed by technical guidance from the Texas Instream Flow Program (TIFP, 2008) but mussels have only been notionally considered and the San Saba was not included in this process. Thus, flow recommendations to mitigate dewatering in Central Texas rivers are likely to be insufficient for protecting mussels, which will further exacerbate conflict between human and wildlife needs in this system.

b. Nature, scope, and objectives of the research

The overall goal of this project is to determine the upper thermal limits of two species of high conservation concern and then relate those findings along with in situ growth studies to water temperature data from perennial and intermittent reaches of the San Saba. This information can then be used to help determine if their decline in this system is related to low flows and elevated water temperature and if so to help inform water management practices that ensure their persistence. The specific objectives of this study are as follows:

Objective 1. Determine lethal response of glochidia and juveniles for select mussel species from the San Saba using standard median lethal temperature (LT50) techniques.

Objective 2. Determine impacts of in situ exposures of juvenile mussels on growth rate and survival within perennial and intermittent reaches of the San Saba River.

Objective 3: To overlay LT50 data compiled from this study against temperature and discharge data along the San Saba to identify E-flow bottlenecks.

Project Timeline

Calendar Year		**2020**			**2021**	
Semester	**Spr**	**Sum**	**Fall**	**Spr**	**Sum**	**Fall**
San Saba River						
Lethal and sublethal effects of temperature (glochidia)		x	x	x		
Lethal and sublethal effects of temperature (juveniles)			x	x	x	
Silo deployment	x					
Silo Collection		x				
Field temperature monitoring	x	x	x	x	x	
Data analysis and thesis preparation				x	x	x

c. *Methods*

To address Objective 1, I will conduct 96-hour static tests (juveniles) and 24-hour static tests (glochidia). Following methods presented in ASTM (2013), juvenile mussels and glochidia will be acclimated to 27˚C and then randomly assigned to one of five different treatment temperatures (30 ˚C, 32 ˚C, 34 ˚C, 36 ˚C, 38 ˚C). These experimental temperatures were chosen based on the range of temperatures mussels encounter within the San Saba River during the warmest months and possible extremes. In addition, an unacclimated control of 20˚C will be examined in comparison to the five experimental treatment temperatures. LT50s, LT05s, and 95% confidence intervals (CIs) will then be determined by a two-parameter regression model using a logistic distribution. The LT50 and LT05 is defined as the temperature that causes mortality in 50% or 5% of the sample, respectively.

To address Objective 2, I will use in situ silos to expose juvenile freshwater mussels to areas of perennial and intermittent reaches within the San Saba River following the methods used in Haag (2019). To determine sensitivity to differences in flow, I will rear *L. bracteata* and *C. petrina* in captivity and allow them to grow out to juveniles in a flow through recirculating system. Silos will be constructed and consist of a concrete dome 26 cm in diameter with a central chamber formed by a 4-inch PVC pipe covered with a drain grate. Mussels will be placed inside of a 3-inch PVC pipe (covered on top and bottom) that will fit within the central chamber and exploit the Bernoulli Principle allowing food and oxygen to be delivered to the mussels and waste to be carried away. Four sites will be identified within the San Saba (two in perennial and two in intermittent reaches), and two silos will be placed at each site. Each of the silos will contain 16 juveniles of one of our two focal species. Juvenile mussels will be grown to a sufficient size and tagged for identification purposes and then measured for both length and weight before being placed in the silos. Mussels will slowly be acclimated to the stream water by gradually replacing the water within the transportation cooler with stream water. Silos will be retrieved after 96 days, and survival and growth rate will be measured and quantified in comparison to stream type (perennial or intermittent).

To address Objective 3, I will deploy data loggers within the San Saba River at the same location where silos are deployed to determine water temperatures at those locations and to further characterize the thermal regime within perennial and intermittent reaches. LT50 and LT05 data from objective 1 will be overlaid on water temperature data to determine if water temperatures are in exceedance of upper limits for my focal species. If this turns out to be the case, then this data will be used to determine the range of discharge during which temperature tolerances are exceeded following methods presented in Castelli et al. (2012).

d. *Statement of expected results or benefits*:

Thermal data of two freshwater mussel species will be obtained and can be used to determine thermal optimums and quantify the responses of freshwater mussel populations in years to come as climate change and more frequent droughts occur. This information can be utilized to develop best water management practices to protect instream flows for both people and wildlife, particularly important to river authorities, municipalities, and other large water users likely to be impacted by the listings.

8. Intended Career Path

I would like to focus my research on the impact of environmental stressors on the physiology of organisms and their ecosystems as well as helping to develop associated conservation policies.

9. Works Cited (not included in 3-page limit)

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