

DESIGN AND FIELD-TEST OF SOLAR-PASSIVE THERMAL
REFUGIA FOR OVERWINTERING MOZAMBIQUE TILAPIA
(*OREOCHROMIS MOSSAMBICUS*) IN CENTRAL TEXAS

A Thesis

by

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ABSTRACT

The aquacultural importance of the Mozambique tilapia (*Oreochromis mossambicus*) as a forage species for largemouth bass (*Micropterus salmoides*) has motivated many private pond owners in the U.S. to try propagating Mozambique tilapia for themselves; however, this species' intolerance to cold poses a challenge for anyone attempting to overwinter these fish outdoors above the Tropic of Cancer. Thus, there is a need for appropriate thermal refuge technology, which still remains unfulfilled. An experimental, solar-passive thermal refuge was designed, produced, and tested in triplicate trials for each of two insulation treatments. The refuge design envisaged a floorless, floating greenhouse with translucent roof and walls. Each unit was made buoyant by a 4" PVC pipe frame attached exteriorly to allow for partial submersion. Walls of three refugia were lined with foam board (Foamular™) to provide additional resistance to heat-flux; whereas, the other three refugia were without foam board. One refuge was installed in each of six similar-sized ponds on the Triple JJJ Ranch of Somerville, Texas, during the winter of 2006-07. Chlorophyll *a* concentrations were generally higher within the refugia than in the ambient water outside, but temperature was not. Although no tilapia survived the winter in this field test, the thermodynamic characteristics of the six floating refugia were monitored and evaluated. A simulation-based analysis indicated that all six refugia afforded substantial thermal inertia, with exponential "decay"-rate coefficients (*k*) ranging from 0.34 to 0.88 °C·day⁻¹·°C⁻¹. The refugia with supplemental insulation actually had larger values of *k* – i.e., less resistance to heat transfer – presumably because they floated higher in the water column, exposing more surface area to the air above and allowing more heat flux beneath due to the increased opportunity for water movement between refuge and pond.

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TABLE OF CONTENTS

	Page
ABSTRACT.....	ii
ACKNOWLEDGEMENTS.....	iii
TABLE OF CONTENTS.....	iv
LIST OF TABLES.....	v
LIST OF FIGURES.....	vii
INTRODUCTION.....	1
MATERIALS AND METHODS.....	5
RESULTS AND DISCUSSION.....	9
CONCLUSIONS.....	20
REFERENCES.....	21
APPENDIX 1.....	25
APPENDIX 2.....	32
APPENDIX 3.....	33

LIST OF TABLES

TABLE	Page
<p>1 Values for area (ha) and depth (m) were measured at the beginning (November 18, 2006) of the study at each experimental and control pond on the Triple JJJ Ranch of Somerville, Texas. Ponds containing refuge with foam board insulation are indicated with an asterisk.....</p>	7
<p>2 Mean ambient temperature, mean refuge temperature, mean differential of temperature (refuge – pond), and median of estimated k [under the assumption that true differential temperature is equal to zero] for all six experimental pond sites on the Triple JJJ Ranch of Somerville, Texas.....</p>	10
<p>3 Values of total hardness (CaCO₃) and alkalinity (CaCO₃) taken at the beginning (November 18, 2006) and end (March 21, 2007) of the study at each experimental pond on the Triple JJJ Ranch of Somerville, Texas. Beginning and ending values were the same for the duration of the study.....</p>	14
<p>4 Ambient and refuge values for chlorophyll a (<i>in situ</i>) within five experimental sites on the Triple JJJ Ranch of Somerville, Texas, for November 30 and December 15, 2006, and March 21, 2007.....</p>	15
<p>5 Mean differentials of temperature and dissolved oxygen (refuge – pond) for each of the refuge units within the six experimental ponds on the Triple JJJ Ranch of Somerville, Texas, from November 20, 2006, to February 14, 2007. Units with foam board insulation are indicated with an asterisk.....</p>	16
<p>6 Ambient and refuge levels of ammonia-nitrogen within four experimental sites on the Triple JJJ Ranch of Somerville, Texas, for December 12, 2006, and March 21, 2007.....</p>	17
<p>7 Median k values, areas, maximum depths, and whether or not supplemental insulation was used for all six experimental pond sites on the Triple JJJ Ranch of Somerville, Texas.....</p>	19
<p>8 Mortality values recorded on December 1, 2006, in regards to time of day, ambient water temperature, refuge water temperature, quantity observed, and individual size range for all nine pond sites on the Triple JJJ Ranch of Somerville, Texas.....</p>	32

9	Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in North J on the Triple JJJ Ranch of Somerville, Texas.....	33
10	Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in South J on the Triple JJJ Ranch of Somerville, Texas.....	33
11	Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Damfoot on the Triple JJJ Ranch of Somerville, Texas.....	34
12	Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Davis Pond on the Triple JJJ Ranch of Somerville, Texas.....	34
13	Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Windmill on the Triple JJJ Ranch of Somerville, Texas.....	35
14	Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Garden Pond on the Triple JJJ Ranch of Somerville, Texas.....	35

LIST OF FIGURES

FIGURE	Page
1 Map of Texas showing the location of Triple JJJ Ranch within the southern post oak savannah ecological region.....	6
2 Ambient versus refuge temperature within Lake North J from November 20, 2006, to February 14, 2007.....	9
3 Ambient versus refuge dissolved oxygen within Lake North J from November 20, 2006, to February 14, 2007.....	16
4 Mean temperature differentials between refugia and associated ponds (Trefuge – Tpond) across all ponds with and without foam board attached to external walls were represented by eight isolated days during the study from January 25 to February 23, 2007.....	18
5 Modeled refuge temperature for Windmill Pond (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = $0\text{ }^{\circ}\text{C}$. Upper panel shows simulated T_r , observed T_r , and observed T_a over the trial; lower panel shows simulated T_r vs. observed T_r , together with the fitted linear regression.....	26
6 Modeled refuge temperature for Damfoot Pond (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = $0\text{ }^{\circ}\text{C}$. Upper panel shows simulated T_r , observed T_r , and observed T_a over the trial; lower panel shows simulated T_r vs. observed T_r , together with the fitted linear regression.....	27
7 Modeled refuge temperature for Davis Pond (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = $0\text{ }^{\circ}\text{C}$. Upper panel shows simulated T_r , observed T_r , and observed T_a over the trial; lower panel shows simulated T_r vs. observed T_r , together with the fitted linear regression.....	28

8	Modeled refuge temperature for Garden Pond (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = $0\text{ }^{\circ}\text{C}$. Upper panel shows simulated T_r , observed T_r , and observed T_a over the trial; lower panel shows simulated T_r vs. observed T_r , together with the fitted linear regression.....	29
9	Modeled refuge temperature for Lake North J (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = $0\text{ }^{\circ}\text{C}$. Upper panel shows simulated T_r , observed T_r , and observed T_a over the trial; lower panel shows simulated T_r vs. observed T_r , together with the fitted linear regression.....	30
10	Modeled refuge temperature for Lake South J (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = $0\text{ }^{\circ}\text{C}$. Upper panel shows simulated T_r , observed T_r , and observed T_a over the trial; lower panel shows simulated T_r vs. observed T_r , together with the fitted linear regression.....	31

INTRODUCTION

The largely tropical fish family Cichlidae includes a group of cichlids, collectively known as tilapia, that are endemic to Africa and are the second most widely farmed freshwater fish in the world (Fitzsimmons 2000). Three genera of aquacultural importance within this group are *Tilapia*, *Sarotherodon*, and *Oreochromis*; however, only members of the genus *Oreochromis*, formerly classified as *Tilapia*, are of commercial importance within the United States today (Rakocy and McGinty 1989). The species within *Oreochromis* of greatest interest in the southern, freshwater sport fishery as quality forage is *Oreochromis mossambicus* (Lock 1988), also known as the Mozambique tilapia; this species is the subject of this study. Other notable members of this genus are the Nile (*O. niloticus*) and the blue (*O. aureus*) tilapias.

The varied diet of the *O. mossambicus* supports the ease of its culture. Natural food organisms consumed include algae, phytoplankton, some macrophytes, zooplankton, benthic invertebrates, larval fish, rooted plants, and detritus (Trewavas 1983, Watanabe et al. 2002). In addition to natural forage, *O. mossambicus* takes readily to pelleted feed. When compared to channel catfish (*Ictalurus punctatus*), 30-50 percent of tilapia growth is attributed to the ingestion of natural food organisms when under a heavy supplemental feeding regimen; whereas, only 5 to 10 percent of channel catfish growth in intensive pond culture is attributable to natural food organisms (Popma and Masser 1999). Natural forage is utilized so efficiently by tilapia that crops of more than 3,000 kg of fish/ha have been cultured in ponds of adequate fertility without supplemental feed; however, *O. niloticus* and *O. aureus* are more efficient at planktonic algal intake than *O. mossambicus* (Popma and Masser 1999).

Tilapia produce more forage as prey than any native fish in Texas (Lock 1988) primarily due to their rapid growth rate, fecundity, and ability to spawn within a relatively wide range of environmental specifications (Siddiqui et al. 1991); consequently, Mozambique tilapia has become a very popular forage species in the trophy largemouth bass (*Micropterus salmoides*) fishery within private impoundments. Mozambique tilapia sexually mature at a younger age than its intrageneric counterparts (Popma and Masser 1999) and possess a high reproductive rate involving six to eight spawns per year; moreover, they reproduce at an even higher rate within smaller ponds—spawning up to

eleven times per year (Pullin and Lowe-McConnell 1982). The number of fry produced per spawn ranges from 250 – 1000 and is positively correlated to the size of the brooding female (Mohamed et al. 2013).

While tilapia are sensitive to low temperatures, they are robustly resistant to other water quality variations that generally have substantial negative impact on other fishes (Chervinski 1982). Tilapia are able to survive at pH ranging from 5 to 11, tolerate nitrite levels up to 89 mg/L for four days, and are more resistant to diseases than many other commonly cultured fish species (Chervinski 1982, Popma and Masser 1999). Although the Mozambique tilapia is relatively resilient to most environmental stressors, it is still vulnerable to un-ionized ammonia toxicity at ammonia nitrogen levels greater than 2 mg/L (Chervinski 1982).

The only major downfall associated with utilizing tilapia as forage is their intolerance to cold temperatures. It has been shown that Mozambique tilapia generally cannot withstand temperatures below 11° C (Watanabe et al. 2002); however, studies have revealed Mozambique tilapia's increased tolerance to low temperature in 5% seawater due to its greater ability to maintain high plasma sodium and chloride concentrations in saline water (Allanson et al. 1971; Chervinski and Lahav 1976; Watanabe et al. 2002). Capacity of cold-tolerance among Mozambique tilapia individuals has been observed to be appreciably variable (Chervinski and Lahav 1976).

In the past, there have been numerous concepts executed in an attempt to overwinter cold-intolerant fishes; success rates varied as well as cost of implementation. Historically, aquaculturists have implemented floating pool covers as thermal blankets over the pond where warm, saline groundwater was pumped beneath (Dorsett 1994), some have constructed greenhouses over the entire pond (Chervinski and Stickney 1980), and others have shifted to intensive culture systems and have moved their cultured organisms indoors for the duration of the cold period (Stickney 1979). In 1991, Michael Schwarz wrote a professional paper on red drum in which he evaluated an experimental thermal refuge that was comprised of a plastic cover positioned over the corner of a pond where heated water was supplied beneath and loosely contained by a pond-ward curtain. Most recently, Dorsett (1994) designed an inflated greenhouse-like structure that was placed over the windward corner of a pond. The inflated structure had heated water

pumped into it allowing for the cultured organisms within the pond to enter and exit the thermal refuge at will (Dorsett 1994). Both Schwarz and Dorsett achieved desired results regarding the function of their refugia. Although Dorsett collected data favoring the thermodynamic efficiency of his refuge, the winters were too mild for him to quantify survival versus mortality among his experiment and controls (Dorsett 1994); however, Schwarz's (unpublished results) effort proved highly effective with no evidence of temperature-related mortality within his experiment over a notably cold winter.

Selection of favorable microhabitat to behaviorally regulate the immediate environmental temperature is the sole means ordinary fish possess to regulate body temperature (Neill 1979). Fish can regulate their metabolism by shifting their location within a thermally heterogeneous environment by means of behavioral thermoregulation (Reynolds and Casterlin 1979; Peterson and Rabeni 1996). Fish movement in response to thermal gradients and discontinuities in lakes (Brandt et al. 1980) and streams (Matthews et al. 1994) and artificial thermal effluent (Neill and Magnuson 1974) has been observed in previous studies. Depending on the mass of the fish, core body temperature can exceed the ambient and peripheral temperatures by a small amount (Reynolds and Casterlin 1979). Most commonly, the fish's body temperature and the occupied ambient water temperature will differ by no more than approximately 0.5-1.0 °C (Lagler et al. 1962; Reynolds et al. 1976).

Undeniably, temperature is a very important component of the aquatic environment, but the totality of the environment plays an even grander role on the fish itself (Neill and Gallaway 1989). The movement and positioning of an individual fish is affected greatly by environmental variables such as temperature, dissolved oxygen, current, and light intensity (Neill and Gallaway 1989). A positive correlation exists between the concentration of dissolved oxygen and the survival time a fish has under lethally high temperatures; moreover, the behavioral response of a fish within a thermal gradient is likely to be strongly influenced by the dissolved oxygen present (Bryan et al. 1984). Therefore, dissolved oxygen determines the potential of a thermally-adequate space becoming an acceptable thermal refuge for fish.

The idea of a cost-effective, solar-passive, and self-sufficient thermal refuge was conceived by Dr. Michael P. Masser, Professor and Extension Specialist at Texas A&M

University and President of the World Aquaculture Society. From his concept, the design of a self-sufficient, floating greenhouse was developed. Following are the essential features of this thermal refuge: The wooden-framed structure is enveloped with twin-walled greenhouse material which would theoretically absorb, transfer, and retain solar energy during daylight hours. The air contained within the refuge would undergo an increase in temperature and would in turn heat the surface of the water. As the water within the refuge stratifies in response to temperature and consequent density differences, the cooler and more dense water would sink out of the open-floored refuge while being displaced by warmer water. The thermal inertia of the water within the insulated refuge is expected to maintain relatively warm temperatures throughout the nights and during cool days. In addition to thermal preservation within the refuge, its greenhouse-like design may also stimulate development of phytoplankton and periphyton within the refuge supplying the tilapia with supplemental nutrients and vitamins (Hargreaves 2000).

The purpose of this study was to develop a practical means of overwintering tilapia in earthen ponds without the use of electricity or an inflow of warm water. The survival of tilapia over the winter via such a solar-passive thermal refuge would enable private pond owners to grow tilapia year-around as bass forage in a relatively simple and economical manner.

MATERIALS AND METHODS

The study area was a 1295 hectare ranch (Triple JJJ) near the town of Somerville, Burleson County, Texas (30.3° N 96.6° W), in the southern post oak savannah region of East-central Texas. Triple JJJ Ranch is approximately ten miles west of the Brazos River and less than one mile north of Yegua Creek. The ranch is drained by perennial, intermittent, and ephemeral creeks. Twenty-eight lakes and ponds ranging from 0.10- 3.70 hectares are located on the property. This region receives 76 - 102 cm of rainfall per year and is characterized by hot, moderately wet summers and cool, dry winters. Extreme temperatures for this region range from -6.6° C in the winter, to 40.5° C in the summer. Rains are sporadic and usually occur most heavily during January and February and from June to August (NWS 2006). Less rain falls between March and May. During my study, July and August were the rainiest months.

The post oak savannah of East-central Texas, within the Texan biotic province, was first described by L.R. Dice (1943), and stretches from the Red River south to the Gulf Coast (Fig. 1). The Kansan, Balconian, and Tamaulipan provinces of grassy, rocky, and semi-arid nature respectively border the Texan province on the west; whereas, the eastern fringe abuts the moist pine-oak forests of the Austroriparian province (Werler and Dixon 2000). Due to its ecotonal nature, the Texan province sustains a fusion of diverse plant and animal species typical of the Tamaulipan and Austroriparian provinces.

Six thermal refugia or floating greenhouses were constructed and placed in six different ponds located on the Triple JJJ Ranch of Somerville, Texas, in the fall and winter of 2006-07 (Table 1). Three additional ponds without refugia were set up as controls. The dimensions of each refuge were 1.85 m by 1.85 m in area and 1.38 m in height. Each refuge consisted of four walls, no floor, and a gabled roof. The exterior was girdled with a four-inch (10.16 cm) diameter PVC pipe frame 60.96 cm up from the bottom edge of the refuge to create buoyancy. When afloat, 61.0 cm of the units' sides remained submerged while the other 75.0 cm, including the gabled roof, was above the water. The internal wooden frame was constructed entirely with untreated two by two lumber (3.81 cm x 3.81 cm) and consisted of a 1.85 m by 1.85 m base frame, corner uprights, a 1.85 m by 1.85 m top frame, joists, and trusses. Heavy wood glue and 5.08 cm wood screws were used to fasten the frames. Each frame was then wrapped and capped

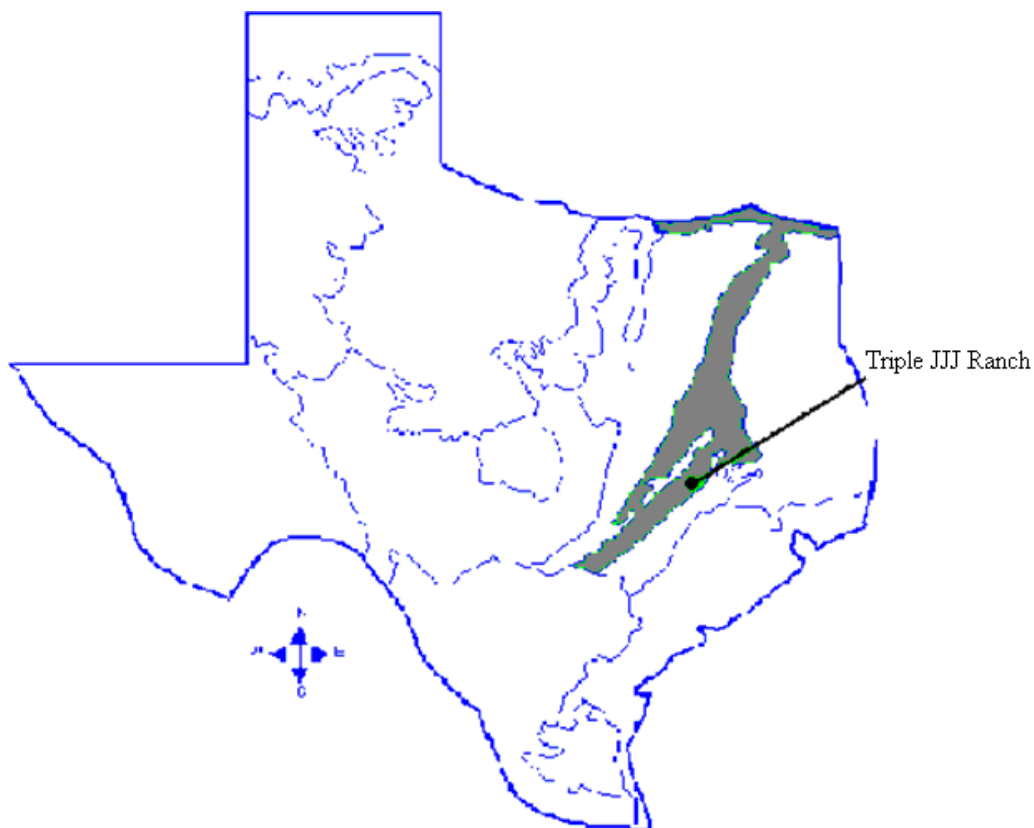


Fig.1. Map of Texas showing the location of Triple JJJ Ranch within the southern post oak savannah ecological region.

with advanced twin-wall greenhouse material (*Solexx*; Adaptive Plastics Inc.), stapled, and caulked with tub and tile silicone. The insulating factor of the twin-wall greenhouse material is a result of its construction, i.e. two walls set apart by a continuous series of horizontal, air-filled channels that measure approximately 5 mm in width. A 4.45 cm diameter PVC pipe was inserted and caulked into each roof peak to provide access for a water quality meter probe and water sample collection. In January of 2007, foam boards (*Foamular*; fiberglass insulation) measuring 60 cm x 185 cm x 1.91 cm were fastened to the four external walls of three refugia in attempt to increase insulation efficacy and generate comparative data.

A YSI 85 meter was used to collect water-temperature and dissolved-oxygen data on 18 different dates throughout the duration of the study. Date, site, and time were also collected for each of the 18 days of measurement. Refuge water samples were collected

via a hand siphon pump and stored in mason jars for subsequent analyses. Refuge and ambient water samples were analyzed for ammonia nitrogen and chlorophyll a (*in situ*). Ammonia nitrogen was measured via *Hach* (Cat. No. 25598-00); Ammonia Nitrogen-Surface Water Quality Test Kit, and a 10-AU *Turner Designs* Fluorometer (*In Situ*) was used to measure concentrations of chlorophyll a. Total hardness and alkalinity were recorded for each pond at the beginning and end of the experiment to ensure the absence of any drastic fluctuations in water chemistry. Total hardness was measured via *Hach*; Total Hardness Titration-Surface Water Quality Test Kit; whereas, alkalinity was quantified via *Hach*; Alkalinity Test Strips.

Table 1

Values for area (ha) and depth (m) were measured at the beginning (November 18, 2006) of the study at each experimental and control pond on the Triple JJJ Ranch of Somerville, Texas. Ponds containing refuge with foam board insulation are indicated with an asterisk.

Pond	Area (ha)	Depth (m)
Lake South J*	0.1	2.15
Damfoot*	0.07	2.46
Davis Pond*	0.06	2.77
Lake North J	0.08	1.54
Windmill	0.07	3.08
Garden Pond	0.08	2.62
Eagle†	0.07	1.85
Blue Lagoon†	0.1	3.7
Deer Pen Pond†	0.08	2.15

Six refuge-equipped ponds as well as three control ponds were stocked with 67.38 kg/hectare of various sized (6-254 mm total length) Mozambique tilapia. The six experimental ponds contained only the tilapia stocked at the beginning of the study, while the three control ponds supported the experimental tilapia in addition to the natural fish assemblages already present at the commencement of the study. All ponds were routinely monitored and controlled for bird depredation on the tilapia.

When measurements of water temperature taken from the inside of each refuge were compared to that of the ambient pond water (Fig. 2) in relation to the duration since the previous measurement at that particular site, an estimation of heat transfer (k) could be determined (Appendix 1). Measurements were taken randomly throughout study period; therefore, time intervals between temperature points were inconsistent (Appendix 3).

At the end of the study, once all six refugia were removed from the ponds, we set out to collect all surviving tilapia by seining the entire area of each pond with a knotless seine measuring 23.11 m in length, 1.85 m in depth, and a mesh of 0.64 cm in size.

Standard deviations of mean differentials for temperature and dissolved oxygen were achieved by means of Statistical Package for the Social Sciences (SPSS).

RESULTS AND DISCUSSION

The overall objective was partly achieved, in that six refuge structures of relatively low cost were developed, deployed, and tested. Material costs for refugia without supplemental insulation and refugia with supplemental insulation were \$675 and \$685 per unit, respectively. The bulk of the cost was associated with the greenhouse material and the 10.16-cm PVC pipe; whereas, the costs for lumber, wood glue, screws, staples, and silicone were minimal. Costs affiliated with the self-sufficient refugia were significantly less when compared to the associated costs of other thermal refuge designs implemented previously, i.e. \$1,150-\$1,200 per unit plus cost of 2.1 million BTU heater (Dorsett 1994). The low-cost nature of these refugia would allow private pond owners to consider implementation of these thermal refugia on an economically practical basis.

Despite all effort put forth into the six experimental ponds equipped with refugia, no tilapia survived the winter in any of the control or experimental ponds; presumably, the mortality was due to lethal cold exposure periodically throughout the relatively mild winter but especially during February 2007 (Weather Warehouse 2013).

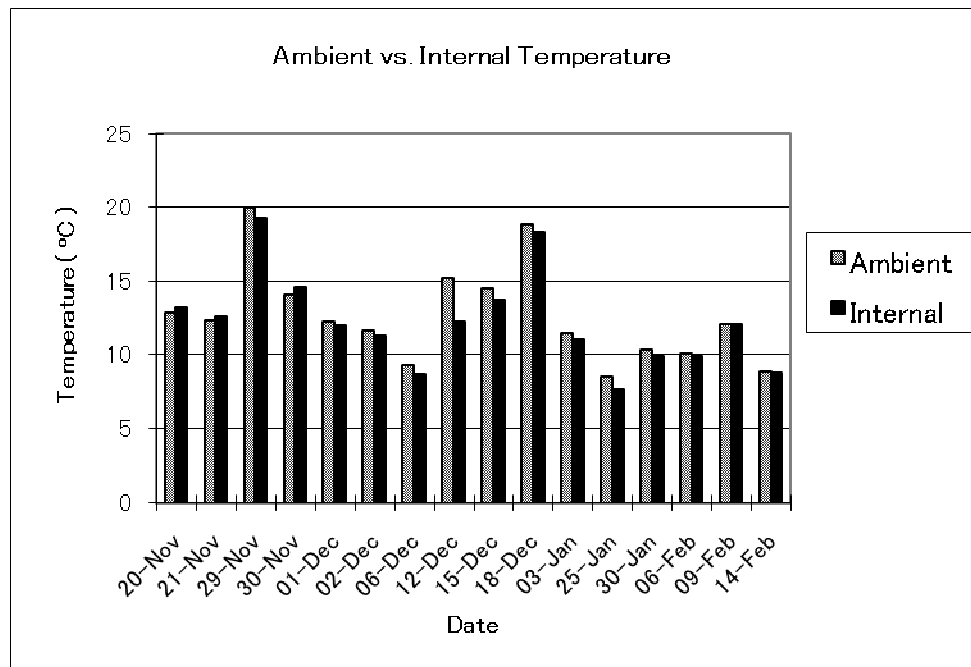


Fig. 2. Ambient versus refuge temperature within Lake North J (without supplemental insulation) from November 20, 2006, to February 14, 2007.

Table 2

Mean ambient temperature, mean refuge temperature, mean differential of temperature (refuge – pond), and median of estimated k [under the assumption that true differential temperature is equal to zero] for all six experimental pond sites on the Triple JJJ Ranch of Somerville, Texas.

Pond	Mean Ambient T °C (SD)	Mean Refuge T °C (SD)	Mean ΔT °C (SD)	Median of Est k 1/day
Lake South J*	13.471 (+/- 3.433)	13.05 (+/- 3.359)	-0.421 (+/- 0.486)	0.693
Damfoot*	12.731 (+/- 3.600)	12.338 (+/- 3.355)	-0.392 (+/- 0.439)	0.843
Davis Pond*	13.546 (+/- 3.313)	12.6 (+/- 3.272)	-1.115 (+/- 1.025)	0.343
Lake North J	11.58 (+/- 3.271)	11.853 (+/- 3.408)	0.273 (+/- 0.193)	0.555
Windmill	13.392 (+/- 3.706)	13.192 (+/- 3.476)	-0.017 (+/- 0.252)	0.576
Garden Pond	13.136 (+/- 3.622)	12.636 (+/- 3.776)	-0.5 (+/- 1.730)	0.883

In general, refuge temperatures “tracked” ambient temperatures, with no evidence of any consistent thermal excess that would indicate solar heating (Table 2). However, simulation modeling suggested substantial thermal inertia, which tended to stabilize refuge temperature in response to abrupt change in ambient temperature. Increased water circulation on windy days may have exacerbated heat transfer between pond and refuge, both through the refuge wall and through the open bottom. An insulated floor with a small opening for entry/exit of tilapia may have reduced heat loss significantly; moreover, increased thermal inertia was observed in units where some part of the refuge bottom was positioned near or in contact with the pond bottom.

The greenhouse material that was employed is not intended for underwater use; therefore, it is presumed that its resistance to conductive and convective heat transfer was minimized when it was submerged in water [due to the differences in thermoregulatory properties of water versus air when the air within the horizontal channels (insulating factor) of the greenhouse material became displaced by water]. The other major contributing factor of heat loss is hypothesized to be due to the absence of a floor or thermal barrier beneath the refuge cavity. On days and/or nights of high winds,

convective mixing may have disrupted stratification and thus destroyed the vertical temperature gradient. Such presumed transient thermocline disruption and consequent mixing would have occurred especially at times when solar energy-input was low or absent, i.e. on overcast days and at night. The effects of the insulating properties of the refugia were both desirable and undesirable. The time period in which the refuge water temperature remained within a favorable range subsequent to a solar event was prolonged due to the high insulating capacity of the greenhouse material. However, when atmospheric air temperature dropped substantially during and/or following a prolonged period of time without direct sunlight (e.g. night), the internal air cooled, and as a consequence the water column within the refuge may have undergone a density turnover leading to the mixing of strata, eventually creating a uniformly lower water temperature throughout the refuge (e.g. as exhibited by refuge data from Lake North J on December 12, 2006: ambient water temperature was 14.9 °C while refuge temperature remained 10.6 °C due to a 3-day arctic cold spell that ended the day prior). Once the cold atmospheric event subsided and the refuge water had its lowest temperature, the insulation seemingly trapped the cold air within the refuge, retarding the subsequent reheating of the water within. At times, the water temperature (e.g. December 12, January 25, January 30) within the refuge remained temporarily lower than that of the ambient water (Fig. 2).

A process cannot be modeled effectively as a simulated exponential decay if the intervals between successive observations of the driving time series are longer than about 2 time constants for the process. In the case of this study, the best-fitting rate constant (k) seemed to be on the order of 0.4/day, which makes the time constant (λ) = $1/0.4 = 2.5$ days. So, when the interval exceeds 5 days, there is really no way to know the extent to which a discrepancy between observed and modeled values of refuge temperature reflects model error versus simple ignorance about what the driving values of ambient temperature were during the period since the preceding observation. The ambient temperature influencing any particular refuge temperature is the value of pond temperature observed at the time when that refuge temperature was observed, and that the pond had that temperature for the entire interval since the previous ambient temperature was observed. It is no surprise that refuge temperature ended up being nearly equal to

ambient temperature when the pond temperature was assumed to be maintained at a constant value for many days, given a time constant of 2.5 days.

Under these circumstances and due to the fish's ability to enter and exit the refuge at will, a fish may exhibit shuttling-type thermoregulatory behavior (Heatwole and Johnson 1979), wherein a poikilotherm strategically centers itself amid a steep temperature gradient in close proximity to the transition line dictated by its physical environment allowing for easy movement to and from specific temperatures. In the case of a floating refuge, as described above, the preferable temperature for Mozambique tilapia may shift from the inside to the outside of the refuge; however, the possibility that the tilapia never utilized or even discovered the refugia may be the reason why no tilapia over-wintered in the refuge-equipped ponds. Not one incident occurred where the experimental tilapia were observed utilizing a refuge; whereas, in a study at Redfish Unlimited in Palacios, Texas, the redfish were observed aggregating within the thermal refuge thus to avoid winter cold-kill (Schwartz unpublished results).

The other, more plausible explanation is that the refugia did not operate as intended to afford the fish actual refuge from lethal cold. Paired environmental data from inside and outside of refugia in the 6 refuge-equipped ponds did not indicate consistent elevation of temperature inside the refugia. In fact, during coldest periods, refuge temperatures were frequently lower than corresponding pond temperatures; thus, even if the tilapia had aggregated inside the refugia, they likely would have achieved no thermal benefit.

Mortality was quantified and recorded on December 1, 2006, and these values are exhibited in Table 8 (Appendix 2). Atmospheric air temperature fluctuated most abruptly between the afternoon of November 29, 2006, and the morning of December 1, 2006; this occurred as a result of the first major cold spell of the year which commenced during the early morning of November 30, 2006. This event was the first to bring the ambient water temperature below the lethal temperature of *O. mossambicus*. Mean ambient water temperature across all experimental ponds dropped from 19.9 °C to 12.2 °C during this 42-hour period.

Direct comparisons of raw temperature data were complemented with more in-depth analysis via simulation modeling of heat-transfer dynamics. This was done to

explore potential benefit of the thermal refugia under conditions different from those observed. The simulation model assumed that refuge temperature (T_r) operated as an exponential filter of ambient water temperature in the pond (T_a), thus obeying “Newton’s Law of Cooling” as adapted by Fechhelm and Neill (1982). This model was fit to the temperature data from each refuge-equipped pond, with and without assuming a steady-state thermal excess.

Simulation confirmed that no substantial temperature-excess existed inside the refugia; therefore, the model with the excess set to 0.0 C was accepted. It indicated that the refugia did have substantial thermal inertia, with values of the temperature-change coefficient ranging from 0.84 to 0.44 day⁻¹. Under conditions of rapidly changing ambient temperature, such thermal inertia could provide substantial protection to fish taking refuge from short-term drops in pond temperature; furthermore, when refuge temperature lagged behind rising pond temperature, fish that had taken refuge from a preceding ambient-temperature drop, need only to leave the “refuge” [which at that point is an anti-refuge] in favor of the open pond, i.e. they need only to exercise the thermoregulatory behavior of which they are very capable.

Initial and final measurements of total hardness (CaCO_3) and total alkalinity (CaCO_3) were taken from each pond in November/March and ranged from 40-220 mg/L and 20-170 ppm, respectively (Table 3). Final measurements were identical to those taken initially implying that the two parameters likely remained relatively constant from the beginning to the end of the study.

Table 3

Values for total hardness (CaCO₃) and alkalinity (CaCO₃) taken at the beginning (November 18, 2006) and end (March 21, 2007) of the study at each experimental pond on the Triple JJJ Ranch of Somerville, Texas. Beginning and ending values were the same for the duration of the study.

Site	Total Hardness (mg/L)	Alkalinity (ppm)
Lake South J	100	110
Lake North J	40	20
Damfoot	220	170
Garden Pond	80	90
Davis Pond	40	35
Windmill	160	80

Concentration of chlorophyll a ($\mu\text{g/L}$) was measured on three dates during the study (November, December, and March) at three random sites per date (encompassing a total of five sites throughout the study) and refuge versus ambient water samples were compared (Table 2). In all but two cases (Lake South J on November 30, 2006, and Damfoot on March 21, 2007), chlorophyll a concentrations within the refugia were higher than that of the ambient water.

A paired samples t-test was performed in SPSS 15.0 to determine whether or not a significant difference existed in chlorophyll a concentrations between refuge and ambient water samples. At the 95% confidence level, paired samples correlations yielded a p-value of 0.001; whereas, the paired samples t-test rendered a p-value of 0.06 (two-tailed). It can be concluded that no difference of statistical significance exists between the refuge and ambient concentrations of chlorophyll a; therefore, the difference between condition means is likely due to chance (Table 4).

Table 4

Ambient and refuge values for chlorophyll a (*in situ*) within five experimental sites on the Triple JJJ Ranch of Somerville, Texas, for November 30 and December 15, 2006, and March 21, 2007. Units with foam board insulation are indicated with an asterisk.

Unit	Chlorophyll a ($\mu\text{g/L}$)- <i>In Situ</i>					
	30-Nov-06		15-Dec-06		21-Mar-07	
	Ambient	Refuge	Ambient	Refuge	Ambient	Refuge
Lake South J*	2.62	2.55	2.03	2.2	--	--
Lake North J	3	4.6	3.5	5.28	1.91	2.04
Garden Pond	2.05	2.39	1.29	1.58	--	--
Windmill	--	--	--	--	2.45	2.71
Damfoot*	--	--	--	--	5.35	4.68

Furthermore, on days with ample sunshine, dissolved oxygen levels within the refugia were at times greater than those of the ambient water (e.g. in Damfoot on November 30 and December 12; Garden Pond on December 1). These two data sets together reveal that oxygen production did occur to some degree within the refugia via algal photosynthesis. Dissolved oxygen levels within the refugia ranged from 3.0-14.3 mg/L with a sample mean of 10.1 mg/L throughout the study; whereas, dissolved oxygen levels among the ponds ranged from 4.1-16.8 mg/L with a sample mean of 10.58 mg/L (Table 5 and Fig. 3).

Table 5

Mean differentials of temperature and dissolved oxygen (refuge – pond) for each of the refuge units within the six experimental ponds on the Triple JJJ Ranch of Somerville, Texas, from November 20, 2006, to February 14, 2007. Units with foam board insulation are indicated with an asterisk.

Unit	Mean Differentials (SD)	
	Temperature (°C)	Dissolved Oxygen (mg/L)
Lake South J*	-0.42 (+/- 0.49)	-0.63 (+/- 0.45)
Damfoot*	-0.39 (+/- 0.44)	-0.07 (+/- 0.05)
Davis Pond*	-1.12 (+/- 1.03)	-0.66 (+/- 0.47)
Lake North J	0.27 (+/- 0.19)	-0.6 (+/- 0.42)
Windmill	-0.02 (+/- 0.25)	-0.5 (+/- 0.35)
Garden Pond	-0.5 (+/- 1.73)	-1.22 (+/- 0.86)

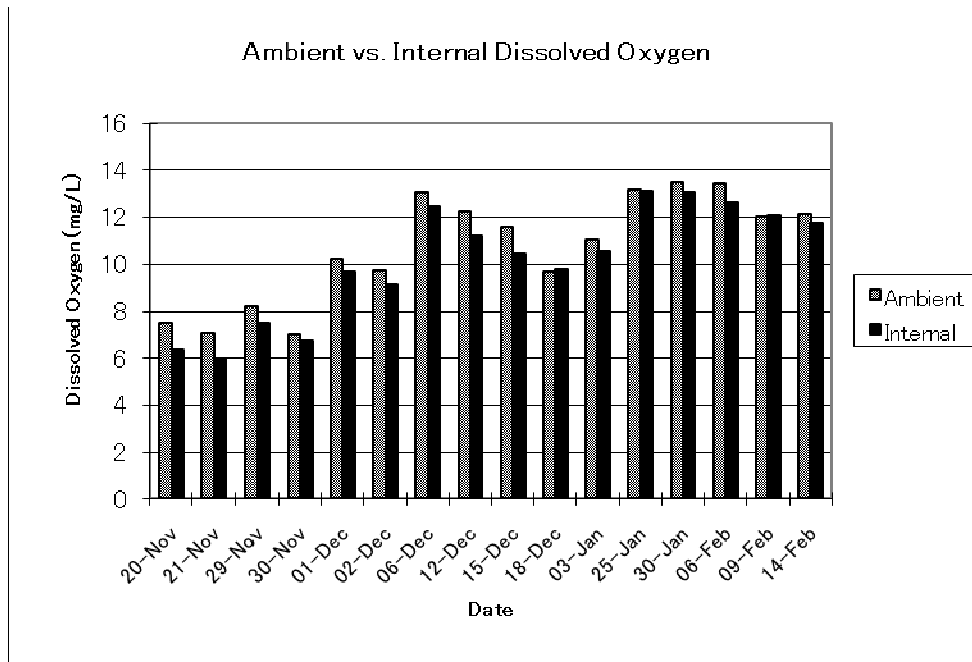


Fig. 3. Ambient versus refuge dissolved oxygen within Lake North J from November 20, 2006, to February 14, 2007.

Un-ionized ammonia (NH₃) nitrogen concentrations were quantified within each pond and refuge in December and March, to evaluate for toxicity potential (Table 6). In December, levels of un-ionized ammonia were all <0.1 mg/L inside and outside the refugia. In March, however, levels had increased to a range of 1.2-1.4 mg/L, but no difference was observed between the refuge and ambient waters in any of the sites. The absence of a significant un-ionized ammonia differential between refuge and surrounding pond water adds support to the hypothesis that substantial water exchange did occur via the open bottoms of the refuges. Un-ionized ammonia nitrogen concentrations no higher than 1.4 mg/L are unlikely to pose a toxicity threat to tilapia (Durborow et al. 1997, Redner and Stickney 1979).

Table 6
Ambient and refuge levels of ammonia-nitrogen within four experimental sites on the Triple JJJ Ranch of Somerville, Texas, for December 12, 2006, and March 21, 2007.

Site	Ammonia-Nitrogen (NH ₃)			
	12-Dec-06		21-Mar-07	
	Ambient	Refuge	Ambient	Refuge
Lake South J	<0.1	<0.1	--	--
Lake North J	<0.1	<0.1	1.4	1.3
Damfoot	--	--	1.3	1.3
Windmill	--	--	1.2	1.2

An improved insulating effect was attempted by adding foam board to the exterior walls of the refugia (Fig. 4); however, the supplemental insulation only increased the undesirable effect of coldwater retention and failed to produce any significant increase in the duration of heat retention. Refugia without supplemental insulation achieved the most desirable results, i.e. longer heat retention and higher observed temperature differentials.

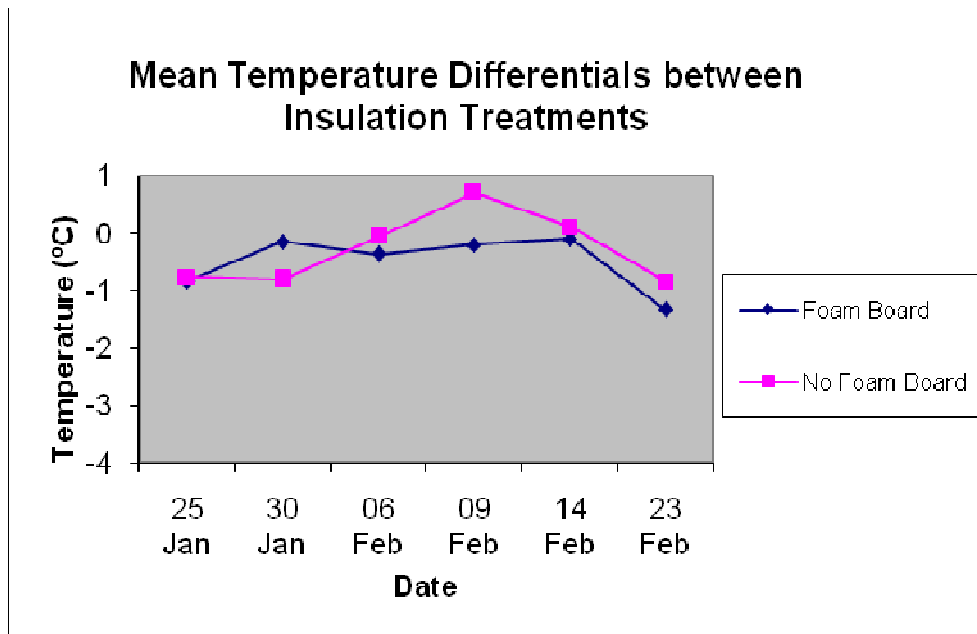


Fig. 4. Mean temperature differentials between refugia and associated ponds (Trefuge – Tpond) across all ponds with and without foam board attached to external walls were represented by eight isolated days during the study from January 25 to February 23, 2007.

One plausible explanation for the unanticipated decrease in efficacy of supplemented refugia may have been due to the foam board’s inherent property of buoyancy. The addition of foam board to three specific refugia increased the buoyancy of the entire units themselves, causing the units to float 10-15 cm higher than those without foam board. This unintentional modification resulted in a smaller volume of water present within each supplemented refuge which likely decreased the thermal inertia of the subjected refugia. The refugia with supplemental insulation had larger values for the heat-transfer coefficient (k) – i.e., less resistance to heat flux – presumably because they floated higher in the water column exposing more surface area to the air above and allowing more heat flux beneath due to the increased opportunity for water movement between refuge and pond.

At the end of the experiment, all refugia were removed from ponds and collection of surviving tilapia was then attempted. Unfortunately (for this project), no fish survived the winter of 2006-07. Differentials in temperature were not significant enough to produce definitive [nor, desired] results. Minimum temperatures recorded for ambient water and refuge water were 7.8°C and 6.9°C, respectively. The maximum observed

positive temperature differential of the refuge water versus the ambient water was 3.4 °C. The maximum observed negative differential between refuge and ambient temperatures ranged from -3.3 °C to -4.3 °C among trials. Of the refugia that exhibited the above stated differentials, only one refuge was equipped with supplemental insulation and only once did it exhibit a significant temperature differential of -3.3 °C; therefore, the added external insulation appeared to have little to no effect (Table 7).

Table 7
Median k values, areas, maximum depths, and whether or not supplemental insulation was used for all six experimental pond sites on the Triple JJJ Ranch of Somerville, Texas.

Site	Area (ha)	Max. Depth (m)	Foam Board	Median k (1/day)
Lake South J	0.1	2.15	Yes	0.693
Lake North J	0.08	1.54	No	0.555
Damfoot	0.07	2.46	Yes	0.843
Garden Pond	0.08	2.62	No	0.883
Davis Pond	0.06	2.77	Yes	0.343
Windmill	0.07	3.08	No	0.576

CONCLUSIONS

An effective and practical means for overwintering tilapia in earthen ponds would benefit private pond owners and aquaculturists alike. Private pond owners would be able to raise tilapia for bass forage or food; whereas, an aquaculturist may be able to produce a similar crop for the commercial market with a substantial decrease in costs. The design described in this thesis is one of the few out there with potential for facilitating this goal. The refuge design evaluated in this study was not successful in the overwintering of tilapia during the winter of 2006-2007, but various modifications to the evaluated design in effort to raise the r-value can be achieved.

The addition of an insulated floor with a small opening for entry/exit to the experimental design evaluated in this study may render a significant decrease in heat loss. Also, preservation of the air within the horizontal channels of the greenhouse material would permit a substantial increase in the r-value of the submerged components; although, the inclusion of a ballast would be necessary to compensate for increased buoyancy.

Even without these improvements, the thermal refuges used in this study had substantial thermal inertia, which could afford some protection from abrupt but short-term cold “snaps,” for cold-intolerant fishes like tilapia.

Time intervals between temperature observations should have been shorter and consistent. The temperature dynamics could have been monitored more precisely throughout the day expressing diurnal fluctuations to allow for determination of a daily average temperature rather than only one isolated observation per day.

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APPENDIX 1

Estimation of the Rate of Heat Transfer (k)

An exponential-decay process was assumed, to describe the dynamic process of heat transfer between water in a refuge and that in the associated pond:

$$\text{SimTr, } ^\circ\text{C} = \text{Tr}_i = \text{Ta}_i - (\text{Ta}_i - \text{Tr}_{i-1}) * \text{EXP}(-k * \Delta t) \quad (1)$$

where Ta_i is pond temperature observed at the i^{th} time; Tr_i (= SimTr, $^\circ\text{C}$) is the corresponding refuge temperature simulated for that time; Tr_{i-1} is the refuge temperature simulated for the just-previous time; Δt is the difference (in days) between the two times, k is the estimated rate coefficient for the negative exponential heat-transfer process, with EXP indicating exponentiation of the napurian (natural) base. All water in the refuge and associated pond at any moment was treated as isothermal, at temperatures Tr_i and Ta_i , respectively.

From the data for each refuge-pond system, median k was computed as the median of the series:

$$k = - \text{LN}((\text{Ta}_i - \text{Tro}_i) / (\text{Ta}_i - \text{Tro}_{i-1}) / \Delta t), \quad (2)$$

where Tro_i and Tro_{i-1} are refuge temperatures actually observed at the i^{th} and the just-previous times, LN indicates natural logarithmic transformation, and other symbols as in equation 1. Logical faults caused by zero denominators and negative arguments of the logarithm (neither of which is allowable under the adopted model) were avoided by setting dis-allowed values arbitrarily to +0.001.

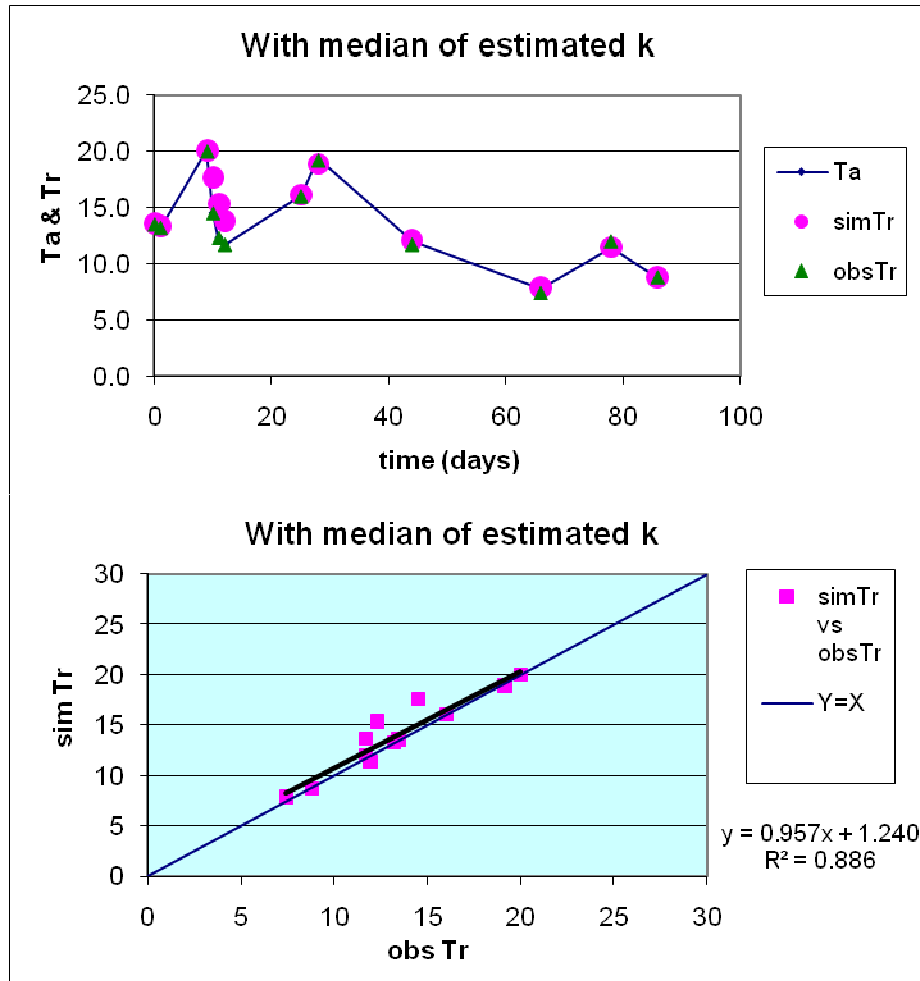


Fig. 5. Modeled refuge temperature for Windmill Pond (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = 0°C . Upper panel shows simulated Tr, observed Tr, and observed Ta over the trial; lower panel shows simulated Tr vs. observed Tr, together with the fitted linear regression.

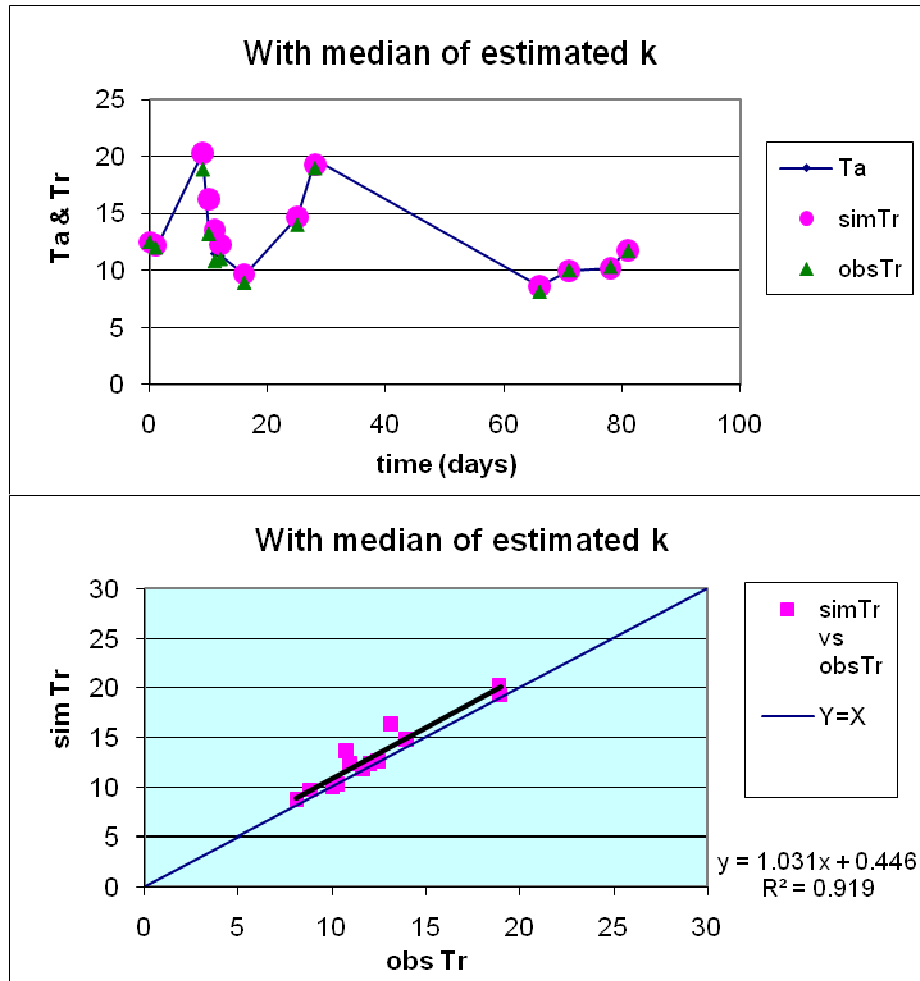


Fig. 6. Modeled refuge temperature for Damfoot Pond (with supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = 0°C . Upper panel shows simulated Tr, observed Tr, and observed Ta over the trial; lower panel shows simulated Tr vs. observed Tr, together with the fitted linear regression.

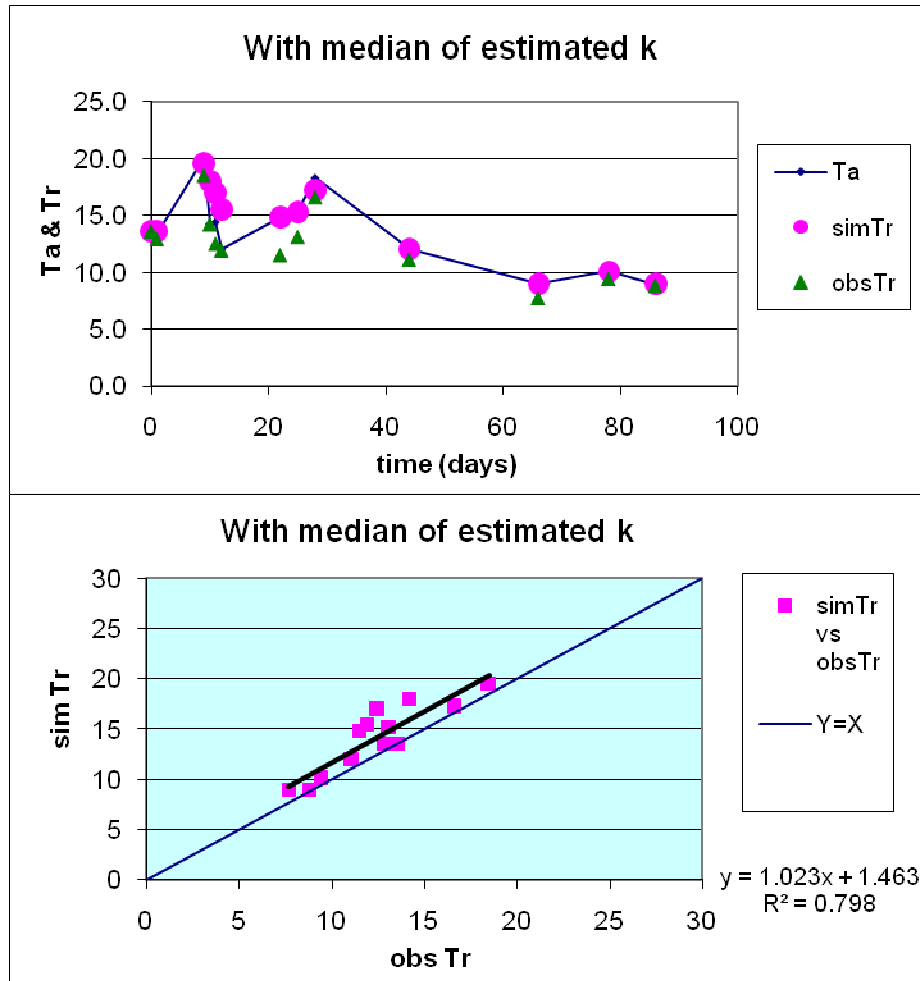


Fig. 7. Modeled refuge temperature for Davis Pond (with supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = 0°C . Upper panel shows simulated Tr, observed Tr, and observed Ta over the trial; lower panel shows simulated Tr vs. observed Tr, together with the fitted linear regression.

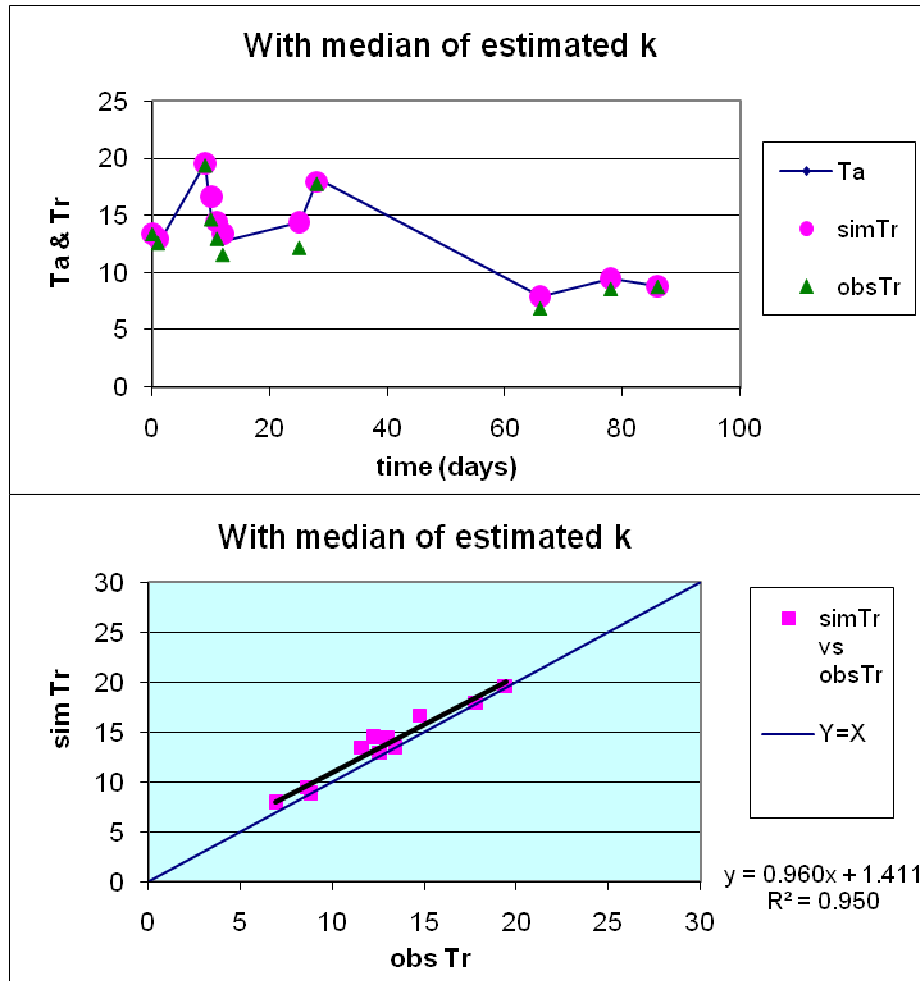


Fig. 8. Modeled refuge temperature for Garden Pond (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = 0°C . Upper panel shows simulated Tr, observed Tr, and observed Ta over the trial; lower panel shows simulated Tr vs. observed Tr, together with the fitted linear regression.

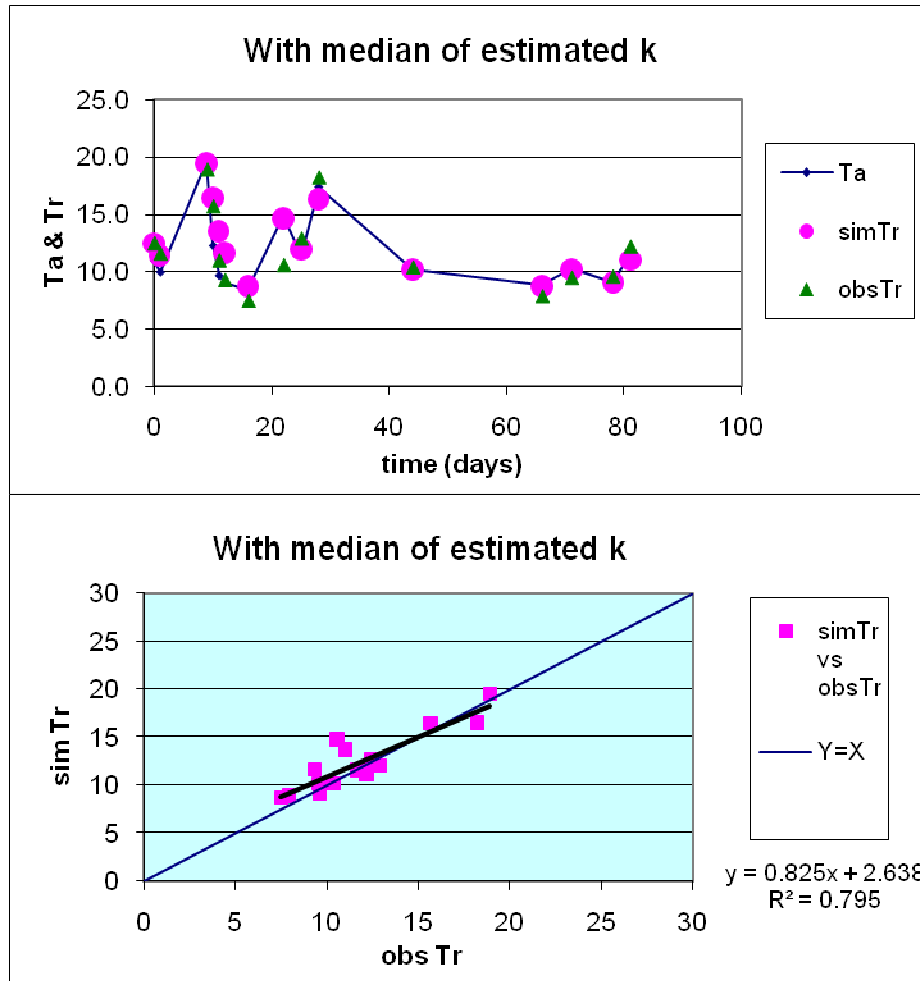


Fig. 9. Modeled refuge temperature for Lake North J (without supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = 0°C . Upper panel shows simulated Tr, observed Tr, and observed Ta over the trial; lower panel shows simulated Tr vs. observed Tr, together with the fitted linear regression.

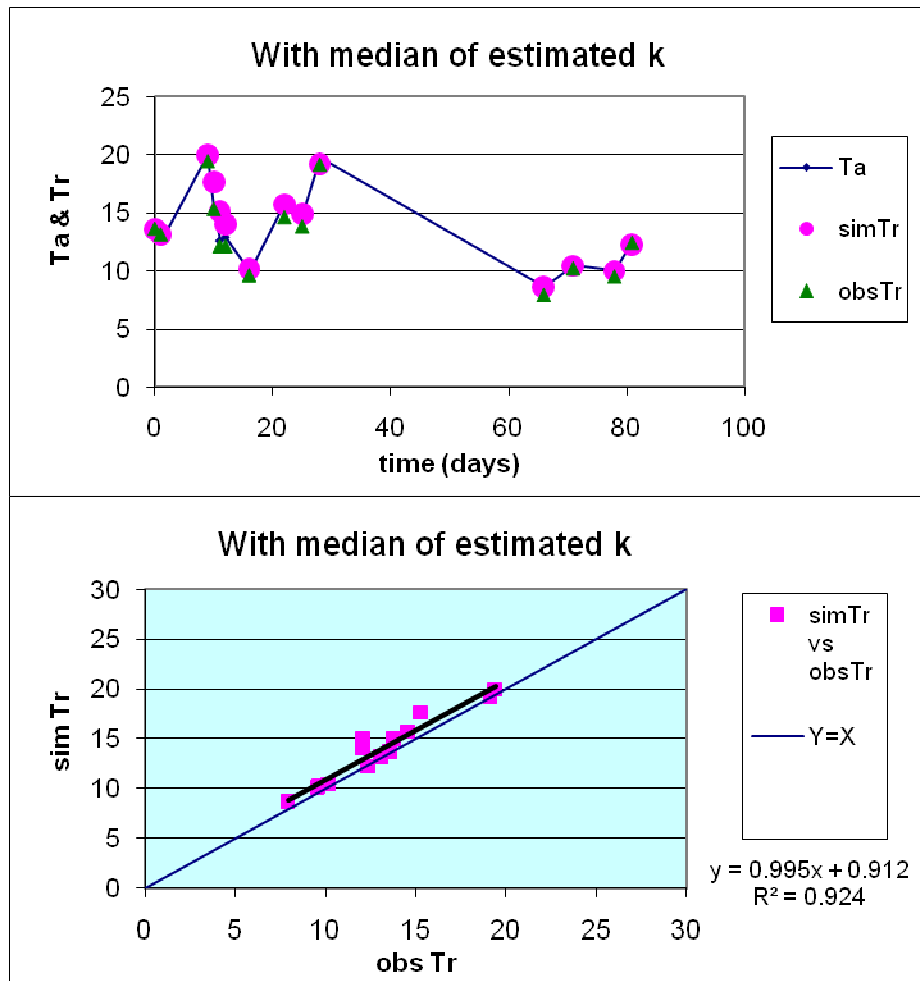


Fig. 10. Modeled refuge temperature for Lake South J (with supplemental insulation), with an estimated median $k = 0.576/\text{day}$, and assuming an equilibrium thermal excess = 0°C . Upper panel shows simulated Tr, observed Tr, and observed Ta over the trial; lower panel shows simulated Tr vs. observed Tr, together with the fitted linear regression.

APPENDIX 2

Table 8

Mortality values recorded on December 1, 2006, in regards to time of day, ambient water temperature, refuge water temperature, quantity observed, and individual size range for all nine pond sites on the Triple JJJ Ranch of Somerville, Texas.

Unit	1-Dec				
	Time	Ambient T °C	Refuge T °C	Quantity	Size (mm)
South J	10:50	12.6	12.1	16	13-210
North J	11:15	9.7	11	151	8-46
Damfoot	11:45	11.5	10.8	27	6-254
Garden Pond	12:15	12.9	13	0	n/a
Davis Pond	12:25	14.4	12.5	0	n/a
Windmill	12:35	12.3	12.3	202	12-210
Eagle†	13:15	11.4	n/a	2	42-54
Blue Lagoon†	13:20	11.7	n/a	1	52
Deer Pen Pond†	13:30	12.5	n/a	0	n/a

APPENDIX 3

Table 9

Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in North J on the Triple JJJ Ranch of Somerville, Texas.

Date	Time	Ambient		Refuge	
		T (°C)	DO (mg/L)	T (°C)	DO (mg/L)
11/20/2006	8:45	11	9.1	12.5	8.2
11/21/2006	9:15	10	8.3	11.6	6.1
11/29/2006	9:20	19.6	8.4	18.9	8.2
11/30/2006	16:30	12.3	9.4	15.7	8.1
12/1/2006	11:15	9.7	13.1	11	11.4
12/2/2006	12:25	9.1	12.2	9.3	11.3
12/6/2006	9:00	8.4	13.8	7.5	13.5
12/12/2006	12:45	14.9	13	10.6	12.2
12/15/2006	9:45	11.4	12.1	12.9	12.3
12/18/2006	9:20	17.4	9.5	18.2	9.7
1/3/2007	12:20	10.2	12.3	10.4	11.7
1/25/2007	12:00	8.8	13.9	7.9	13.6
1/30/2007	15:00	10.3	13.8	9.5	13.2
2/6/2007	9:20	9.1	12.9	9.6	13.1
2/9/2007	14:25	11.5	12.4	12.2	12.6

Table 10

Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in South J on the Triple JJJ Ranch of Somerville, Texas.

Date	Time	Ambient		Refuge	
		T (°C)	DO (mg/L)	T (°C)	DO (mg/L)
11/20/2006	8:30	13.2	9.3	13.6	7
11/21/2006	9:05	12.6	6.7	13.1	6.3
11/29/2006	9:05	20	7.3	19.4	6.6
11/30/2006	16:10	15.3	6.9	15.3	6.7
12/1/2006	10:50	12.6	9.9	12.1	9.5
12/2/2006	12:50	12.9	9.4	12.1	8.7
12/6/2006	8:50	9.9	11.6	9.6	10.9
12/12/2006	12:20	15.8	11.6	14.6	11.2
12/15/2006	9:15	14.8	11.1	13.8	10.1
12/18/2006	9:10	19.8	10	19.1	9.2
1/25/2007	11:50	8.6	12.8	7.9	13.2
1/30/2007	14:40	10.5	12.9	10.2	12.2
2/6/2007	9:00	10	11.5	9.5	10.6
2/9/2007	14:10	12.6	10.6	12.4	10.6

Table 11

Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Damfoot on the Triple JJJ Ranch of Somerville, Texas.

Date	Time	Ambient		Refuge	
		T (°C)	DO (mg/L)	T (°C)	DO (mg/L)
11/20/2006	9:05	12.5	8.8	12.5	8.2
11/21/2006	9:55	12	7.5	12	7.2
11/29/2006	10:00	20.3	8.3	18.9	7.6
11/30/2006	16:45	13.2	8.6	13.2	9.2
12/1/2006	11:45	11.5	12.7	10.8	12.4
12/2/2006	13:15	11.3	12.1	11	11.9
12/6/2006	9:15	9.6	13.8	8.9	12.9
12/15/2006	10:10	14.7	12.9	14	13
12/18/2006	9:35	19.7	11.6	19	13.6
1/25/2007	12:15	8.6	13.9	8.1	13.7
1/30/2007	15:30	10	13.8	10	13.8
2/6/2007	9:30	10.2	14	10.3	13.7
2/9/2007	14:35	11.9	13	11.7	12.9

Table 12

Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Davis Pond on the Triple JJJ Ranch of Somerville, Texas.

Date	Time	Ambient		Refuge	
		T (°C)	DO (mg/L)	T (°C)	DO (mg/L)
11/20/2006	10:35	13.4	5.6	13.5	5.5
11/21/2006	9:40	13.1	7.2	13.2	6.4
11/29/2006	10:15	20.1	8.7	20	8.7
11/30/2006	17:45	14.5	7.2	14.5	7
12/1/2006	12:25	14.4	9.3	12.5	8.1
12/2/2006	14:10	12	8.3	11.9	8.1
12/12/2006	13:45	14.8	12.1	11.5	10.2
12/15/2006	10:40	15.5	11.6	13.1	10.7
12/18/2006	9:55	18.3	10.3	16.6	9.6
1/3/2007	13:00	12	10.5	11.1	9.5
1/25/2007	12:30	9	12	7.7	11.9
2/6/2007	9:40	10.1	14.2	9.4	13
2/14/2007	17:30	8.9	11.6	8.8	11.3

Table 13

Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Windmill on the Triple JJJ Ranch of Somerville, Texas.

Date	Time	Ambient		Refuge	
		T (°C)	DO (mg/L)	T (°C)	DO (mg/L)
11/20/2006	9:20	13.6	6.6	13.6	5.5
11/21/2006	10:15	13.5	7.1	12.9	5.3
11/29/2006	9:45	20	9.3	18.5	8.5
11/30/2006	17:05	14.2	5.8	14.2	5.6
12/1/2006	12:35	12.3	10.5	12.3	10.3
12/2/2006	14:20	11.7	9.8	11.7	9.4
12/15/2006	10:55	16.1	13.8	16	13.5
12/18/2006	10:20	19.4	11.1	19.2	10.6
1/3/2007	13:20	12	10.2	11.7	10.3
1/25/2007	12:50	7.8	13.6	7.4	13.5
2/6/2007	10:05	11.4	14.7	12	14.3
2/14/2007	17:55	8.7	13.6	8.8	13.3

Table 14

Ambient and refuge values for temperature and dissolved oxygen in relation to date and time of day in Garden Pond on the Triple JJJ Ranch of Somerville, Texas.

Date	Time	Ambient		Refuge	
		T (°C)	DO (mg/L)	T (°C)	DO (mg/L)
11/20/2006	10:20	13.3	5.6	13.4	3.7
11/21/2006	10:05	12.6	5.5	12.6	4.4
11/29/2006	9:35	19.6	7.2	19.4	5.5
11/30/2006	17:20	14.6	4.1	14.7	3.8
12/1/2006	12:15	12.9	5.6	13	6.3
12/2/2006	13:30	12.7	6.4	11.6	5.2
12/15/2006	10:25	14.4	7.8	12.2	3
12/18/2006	9:45	18.2	5.4	17.8	5.7
1/25/2007	12:40	7.9	12.7	6.9	12.6
2/6/2007	9:50	9.5	13.4	8.6	10.8
2/14/2007	17:40	8.8	11.2	8.8	10.5