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# Investigation of the Enviromental Factors Associated With the Temporal Abundance of Lutzomyia Anthophora on a Ranch Near Poth, Texas

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# INVESTIGATION OF THE ENVIRONMENTAL FACTORS ASSOCIATED WITH THE TEMPORAL ABUNDANCE OF *LUTZOMYIA ANTHOPHORA* ON A RANCH NEAR POTH,TEXAS

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# INVESTIGATION OF THE ENVIRONMENTAL FACTORS ASSOCIATED WITH THE TEMPORAL ABUNDANCE OF LUTZOMYIA ANTHOPHORA ON A RANCH NEAR POTH,TEXAS

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Leishmaniasis is caused by infection with protozoan parasites within the genus *Leismania* transmitted by the bites of female sand flies within the genus *Lutzomyia* in the New World. This study investigated the seasonal abundance of sand flies and evaluated the correlation of temperature with abundance data. Sand flies were collected at the Garrison ranch near Poth, Texas (28° 57' 10" N; 98° 7' 28" W) from May in 2014 through October, 2015. Recorded highest abundance of sand flies in 2014 was in October, 2 females *Lu.anthophora* and 5 males *Lu.anthophora* were captured producing 0.58 sand flies /trap night, while, mean ambient air temperature was 23.35°C, and mean temperature of the woodrat nest was 23.57°C. In 2015, recorded highest abundance of sand flies was in July, 7 female and 4 males *Lu. anthophora* were captured, producing 0.91 sand flies /trap night. While mean ambient air temperature was 29.17°C, and mean temperature of the woodrat nest was 27.74°C. during the given data of 18 months, strongest positive correlation with ambient air temperature and woodrat nest temperature and sand fly abundance. Increased sand fly abundance was associated with an increase in ambient air temperature and woodrat nest temperature. In Texas small changes in temperature can expand the geographical area in which the *Leishmania* is able to replicate, allowing increase in endemic areas.

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# LEISHMANIASIS

The term leishmaniasis designates a collection of diseases that are caused due to infection by the protozoan parasite *Leishmania* (genus). Leishmaniasis is primarily a vector borne zoonotic infection, which includes animal reservoir hosts and sandflies in the transmission cycle. This is a disease of poverty as its victims are among the poorest. According to ranking, after malaria, it is the second most prevalent parasitic disease. Leishmaniasis causes considerable morbidity and mortality. This disease has traditionally been classified in three major forms on the basis of clinical symptoms; these are cutaneous, muco-cutaneous and visceral leishmaniasis. In cutaneous leishmaniasis, parasites stay near the area of the sand fly bite, while in mucocutanea leishmaniasis parasites metastasize to the mucosa of the nose, mouth, and throat. In visceral leishmaniasis parasites metastasize to internal organs, most commonly the liver and spleen. The most deadly form is visceral, which if left untreated leads to death (Desjeux 1996; Handman 2001; Janovy et al. 2009).

Leishmaniasis is more common in tropical and subtropical climates, and occurs on all continents except Antarctica. High-risk environments for the disease include arid, rural areas, tropical forests, canyons and urban environments. (Neghina R and Neghina M N 2010).

Leishmaniasis is a vector-borne disease transmitted by sand flies within the genus Lutzomyia in the New World and Phlebotomus in the Old World. Leishmaniasis is chiefly spread from animals to human, which is why it is considered a zoonotic disease with diverse reservoir hosts including wild foxes, dogs, and rodents; humans are accidently infected, moreover, humans also aid as a parasitic reservoir, mainly throughout epidemics of disease. The protozoan is transmitted to

humans through the bite of an infected female sand fly from the genus *Phlebotomus* or *Lutzomyia*. However, incidental transmission by blood transfusions or by needle sharing among intravenous drug addicts has also been described (Desjeux 1996; Dostálová and Volf 2012; Janovy et al. 2009).

Leishmania has a dimorphic life cycle. The life cycle begins with promastigotes that are present in sand flies. The promastigote is the extracellular, motile body form with an anterior flagellum at the end of the cell representing the first replicative forms that multiply by longitudinal binary fission in the sand fly and are separated from the midgut by Type-I peritrophic matrix. When the sand fly bites, it injects promastigotes into humans or any other mammal that is susceptible to the disease. When these promastigotes enter the bloodstream of humans, the immune system activates, producing macrophages, which ingest the promastigotes; after ingestion these promastigotes transform into amastigotes. The amastigotes are without a flagellum and are the intracellular, round in shape and immobile form within the vertebrate host. It lives and reproduces asexually inside a phagolysosome of macrophages. The amastigotes multiply and increase their numbers, until the macrophage bursts. They then infect other macrophages where asexual reproduction continues. The skin is the permanent site for amastigotes during cutaneous leishmaniasis. On the other hand, the complete reticuloendothelial system is under attack during visceral infection. If the patient is again bitten by a sand fly and it takes up a blood meal then the life cycle is completed (Handman 2001; Chappuis et al. 2007).

Leishmania promastigotes escape from host immune system through various mechanisms. In order to survive the harsh conditions within the phagosome such as

lower pH, temperature extremes, and increased oxidative and nitrosative stress, promastigotes are internalized into endosomal compartments. These promastigotes prevent fusion of the phagosome and lysosome thus, inhibiting the endosomal maturation. The extracellular surface of Leishmania is rich in lipophosphoglycans (LPGs), and the parasites secrete a large number of peptidophosphoglycans and glycoproteins whose role is not completely understood. Accumulation of periphagosomal F-actin is induced by LPG, which inhibits the endosome maturation. Lipophosphoglycans also prevent acidification of the phagosome by interfering with the V-ATPase pump. Lipophosphogly ontrols the acidification of phagosome that enables the promastigotes to transform into resistant amastigotes (Roberts, Janovy and Schmidt, 2009).

I investigated the seasonal abundance of sand flies at a ranch near Poth,Texas. Analysis of sand fly prevalence and the associated environmental factors were then evaluated to determine if correlation exists between sand fly abundance, ambient temperature, and woodrat nest temperature

# LITERATURE REVIEW

## *Distribution of Leishmaniasis in the New World*

Leishmaniasis occurs focally in the Americas from Yucatan, Mexico, through central and South America to the Peruvian Andes. Different species are named and grouped based on the type of disease that they primarily cause. Cutaneous leishmaniasis is the most common form of leishmaniasis affecting humans caused by *Leishmania mexicana* in the New World. The parasites stay near area of sand fly bite. In mucocutaneous leishmaniasis the parasites infect the mucosa of nose, mouth, and throat. Mcocutaneous leishmaniasis is caused by *Leishmania braziliensis*. In visceral leishmaniasis the parasites migrate to the internal organs such as the liver and spleen primarily is caused by *Leishmania chagasi* (Desjeux, 1996; Handman, 2001; Roberts, Janovy and Schmidt, 2009).

## *Leishmaniasis in Texas*

Several foci of endemic leishmaniasis have been reported in the United States (Gratz, 2006;Mchugh et al., 2001). In the United States, autocthonous human cutaneous leishmaniasis caused by Leishmania mexicana appears to be limited to south central Texas (Ryan et al., 2012). The first autochthonous case of cutaneous leishmaniasis was reported near the Mexican border of Texas in 1903 (Alvar et al., 2012). Cutaneous leishmaniasis has also been found in northern areas of Texas and may be associated with sand flies in some cases. It is noted that the disease is generally endemic to south-central Texas, where it is associated with rodent hosts. Nine autochthonous cases have, however, been reported in northern areas of the state despite patients having no previously reported travel to areas endemic for leishmaniasis (Wright, et al. 2008).

*Sand flies and seasonal abundance in Texas*

McHugh, et al. (2001) collected sand flies from Medina County and Bexar County in Texas, between April and October, was suggesting a warmer climate element to the disease. In the study by Mchugh et al. (2001) the sand fly species *Lutzomyia diabolica* and *Lutzomyia anthophora* were collected over periods of several months in Texas and analyzed in terms of their seasonal abundance. The sampling duration in Medina County was from April until October in the year 1997. The samples in Bexar County were collected over a year from April 1998 till December 1999. Of the two species studied, *Lu. diabolica* samples were obtained from April through November. The highest abundance was witnessed in July. On the other hand, *Lu. anthophora* samples were obtained from April through November, the highest abundance was witnessed in September. The researchers witnessed three generations during the duration of study. The occurrence of three generations were marked by three peaks of abundance in July. Another species of sand fly studied was *Lutzomyia texana* .These were also collected over a span from April through October in 1997. The number of these sand flies increased and reached a maximum abundance in April during 1997(McHugh et al., 2001).

*Reservoir hosts in Texas*

White-throated rats (*Neotoma albigula*) infected by *L. mexicana* were identified near Tucson Arizona in 1998 (Kerr et al., 1999). An eastern woodrat (*Neotoma floridana*) infected by *L. mexicana* was identified in Medina County, Texas (McHugh et al., 2001). It is well documented that *Leishmania mexicana* is the major cause of cutaneous leishmaniasis in cats in Texas. The local vectors for *Leishmania mexicana* are *Lutzomyia diabolica* (anthropophilic) and *Lu.anthophora* (zoonotic) (Clarke et al., 2013; C. P. McHugh et al., 2001). *Lutzomyia anthophora* and *Lu.*

*diabolica* are the species of the sand flies supposed to be involve in the transmission of the parasite to humans (González et al., 2011; C. P. McHugh et al., 1996). So, the vector for *L. mexicana* in humans and mice are *Lu. diabiolica* and *Lu. anthophora*, respectively (Endris et al., 1987; Lawyer et al., 1987). In addition, *Neotoma micropus* is a zoonotic reservoir of *Leishmania* in Texas, and has been proven to be the basic host. When an infected rodent is bitten by *Lutzomyia anthophora* the parasite is ingested with their blood then people are infected if bitten by the same sand fly. So leishmaniasis is commonly associated with the distribution of *Neotoma micropus*. The risk factors involved include living or working in a woodland habitat (McHugh et al., 1996). The majority of these cases were recorded in the cooler months of the year, suggesting a different ecological pattern for the disease in rodent hosts (Raymond et al., 2003).

### *Sand fly abundance in Texas and environmental factors*

The sand flies are usually found near water so that the females can lay eggs either in damp soil or in water the life cycle of sand flies generally begins in the start of spring and may last till the end of June however, in very warm regions like South Texas the adult activity may continue the whole year. A study by Gonzalez et al, (2010) suggested that a change in climate towards warmer temperature would put populations at increased risk for contracting leishmaniasis. Almost all the species cause a painful bite and some of them transmit diseases. Most of the species are crepuscular; they are active at sunset and sunrise (Hernández-Torres et al.,2015).

However, some species are diurnal, known to be active during the whole day and some species are nocturnal. Both the male and female sand flies commonly the fed on juices of leaves and flowers, while females of most of the species require blood

intake egg development. Mainly they are bloodsuckers of vertebrates, including mammals and birds. Others feed on blood of larger insects like moths and dragon flies. Their mouth parts act in a saw like motion to enter through the skin which causes an intense pain leaving a red dot on the spot and itching that can lasts for several days (Jeziorski et al.,2015).

The enhancement of seasonal flies may occur specifically in spring to summer in Texas but may spread over the whole year to warmer areas like south Texas (Kumari et al. 2015). Also marshy areas are supposed to hold major and dense population of sand flies. Some sand flies species are also seem to be associated with presence of forests. The sand flies lay eggs in batches of 30-40, forty up to 450 in total (Kumari et al., 2015).

The larvae move in soil, primarily in coastal or watery areas. Some species show a pause in larvae development when encounter a dry land and soil and resume their growth on arrival of seasonal rains. Adults typically prefer to live in humid and marshy areas. Some species of sand flies are common in coastal salt marshes and inland saline wetlands (Nzelu et al., 2015).

# MATERIALS AND METHODS

## *Sand fly collection and identification*

Field research was conducted at the Garrison ranch near Poth, Texas (28° 57' 10" N; 98° 7' 28" W). The trapping site is primarily characterized by mesquite trees (*Prosopis glandulosa*), and prickly pear cactus (*Opuntia sp*), the soil type is Clay loam (united states. Bueau of soils map,Texas,Wilson county, map, June 9, 1908)

Sand flies were collected using Hoch New Standard Miniature Light traps (John W. Hoch Company, Gainesville Florida). These traps contained a light, a fan and a collection reservoir. The traps had a photo sensor that activates the light at dusk and turns it off at daybreak in order to conserve battery power for the fan. Insects are attracted to the light and the fan forces them into the collection reservoir. Traps were set close to the nests of the Southern plains woodrat (*Neotoma micropus*) during the day and collected the next morning.

The insects were returned to the laboratory and the collection reservoirs were placed in a ⁻20° C freezer for 30 minutes to kill the insects. The insects were then placed in a Petri dish, and the sandflies were sorted from the other insects using a dissecting microscope. The sand flies were then placed in a 500µL microcentrifuge tube containing 70% isopropanol and vortexed for 20 seconds. This was done to remove excess hairs from the sand flies. The sand flies were then placed on a microscope slide and sexed. The males were identified to species under a microscope using the guide of Young and Duncan (1994). Males were identified by the structure of the terminalia. The females were placed on a microscope slide, dissected by removing the last two body segments. The females were identified to species by

observing the structure of the spermatheca and using the guide of Young and Duncan (1994).

*Collection of environmental data*

Using a Davis Vantage Pro2 (Davis Instruments Corporation, Hayward, California) weather station ambient temperature data was recorded. A temperature probe from the station was placed in a woodrat burrow to record nest temperature.

*Analysis of sand fly prevalence and environmental data*

Monthly sand fly abundance data then were compared with ambient air temperature and nest temperature and evaluated for correlation using SPSS software and Pearson's product-moment correlation. Monthly sand fly abundance was converted to sand flies per trapnight in order to normalize the data. Analysis of correlation of sand flies per trapnight was accomplished against the following variables: monthly maximum ambient temperature, monthly minimum ambient temperature, monthly average ambient temperature, monthly average ambient daily high temperature, monthly average ambient daily low temperature, monthly maximum nest temperature, monthly minimum nest temperature, monthly average nest temperature, monthly average nest daily high temperature, and monthly average nest daily low temperature.

A value of Pearson's Correlation Coefficient of + 0.1 to + 0.3 shows a weak positive linear relationship; whereas correlation of - 0.1 to - 0.3 shows a weak negative linear relationship. A value of correlation of + 0.3 to + 0.5 shows a moderate positive linear relationship; whereas correlation of - 0.3 to - 0.5 shows a moderate negative linear relationship. Lastly a value of correlation of + 0.5 to + 10 shows a strong

positive linear relationship; whereas correlation r - 0.5 to - 1.0 shows a strong negative linear relationship.

# RESULTS

## *Temporal abundance of sand flies in 2014*

During the period May through December 2014, eighteen trap nights produced a total of 12 sand flies. The months of May, June, November and December produced zero sand flies. In July, 1 male *Lu. anthophora* was captured, producing 0.3 sand flies/trap night. In August, 2 female and 1 male *Lu. anthophora* were captured, producing 1 sand fly/trap night. In September, 1 female *Lu.anthophora* was captured producing 0.3 female sand flies/trap night. In October, 2 female *Lu.anthophora* and 5 male *Lu.anthophora* were captured producing 0.58 sand flies/trap night. The highest abundance in 2014 of 7.0 *Lutzomyia anthophora* was witnessed in October. (Table 1) *Ambient Air Temperature and Woodrat Nest Temperature in 2014*

The highest average monthly ambient air temperature occurred during August and was 30.21°C. The lowest average monthly ambient air temperature occurred during December and was 10.08 °C. The highest average monthly woodrat nest temperature occurred during August and was 29.62°C. The lowest average monthly woodrat nest temperature occurred during December and was12.53°C (Figure 1)

The highest maximum ambient air temperature of 40.5°C occurred in August. The minimum ambient air temperature of ¯2.3°C occurred in December. The maximum woodrat nest temperature of 33.3°C occurred in August and September. The minimum woodrat nest temperature of 6.1°C occurred in December. (Figure 2)

The maximum average ambient air daily temperature of 38.6°C occurred in August. The minimum average ambient air daily temperature of 3.4°C occurred in December. The maximum average woodrat nest daily temperature of 31.5 °C occurred in August. The minimum average woodrat nest daily temperature of 9.7°C occurred in December. (Figure 3)

*Temporal abundance of sand flies in 2015*

During the period January through October 2015, Ninety-one nights produced a total of 28 sand flies. The months of January, February, March, April, May, and June produced zero sand flies. In July, 7 female and 4 male *Lu. anthophora* were captured, producing 0.91 sand flies /trap night. In August, 1 female *Lu. anthophora* and 2 male *Lu .anthophora* and 2 male *Lu. texana* were captured, producing 0.41 sand flies /trap night. In September, 3 female and 4 male were captured, producing 0.58 sand flies /trap night. In October, 5 male of *Lu.anthophora* were captured, producing 0.83 sand flies /trap night. Highest abundance in 2015 of 11.0 *Lu.anthophora* was witnessed in July. (Table 1)

*Ambient air temperature and woodrat nest temperature in 2015*

The highest average monthly ambient air temperature occurred during August and was 29.69° C. The lowest average monthly ambient air temperature occurred during January and was 8.80° C. The highest average monthly woodrat nest temperature occurred during September and was 29.42° C. The lowest average monthly woodrat nest temperature occurred during January and was 10.38° C. (Figure 1)

The highest maximum ambient air temperature of 40.6° C occurred in August. The minimum ambient air temperature of ¯1.4 ° C occurred in January. The highest maximum woodrat nest temperature of 32.2° C occurred in August and September. The minimum woodrat nest temperature of 5.6 ° C occurred in January. (Figure 2)

The maximum average ambient air daily temperature of 38.1° C occurred in August. The minimum average ambient air daily temperature of 2.8° C occurred in January. The maximum average woodrat nest daily temperature of 31.0 °C occurred

in September. The minimum average woodrat nest daily temperature of 8.5°C occurred in January. (Figure 3)

Table 1
*Temporal Abundance of Sand Flies in 2014-2015*

| Months | Sand flies male | Sand flies female | Average monthly Temperature ambient air | Average monthly Temperature woodrat nest |
|---|---|---|---|---|
| May 2014 | 0 | 0 | 25،48 | 24،14 |
| Jun2014 | 0 | 0 | 28،10 | 26،74 |
| Jul2014 | 1 | 0 | 28،60 | 27،09 |
| Aug2014 | 1 | 2 | 30،21 | 29،62 |
| Sep2014 | 0 | 1 | 26،16 | 27،01 |
| Oct2014 | 5 | 2 | 23،35 | 23،57 |
| Nov2014 | 0 | 0 | 15،11 | 17،45 |
| Dec2014 | 0 | 0 | 10،08 | 12،53 |
| Jan2015 | 0 | 0 | 8،80 | 10،38 |
| Feb2015 | 0 | 0 | 11،24 | 13،05 |
| Mar2015 | 0 | 0 | 15،92 | 15،40 |
| Apr2015 | 0 | 0 | 21،65 | 21،44 |
| May2015 | 0 | 0 | 23،98 | 23،21 |
| Jun2015 | 0 | 0 | 26،72 | 26،37 |
| Jul2015 | 4 | 7 | 29،17 | 27،74 |
| Aug2015 | 4 | 1 | 29،69 | 29،18 |
| Sep2015 | 4 | 3 | 28،72 | 28،72 |
| Oct2015 | 5 | 0 | 23،13 | 25،01 |

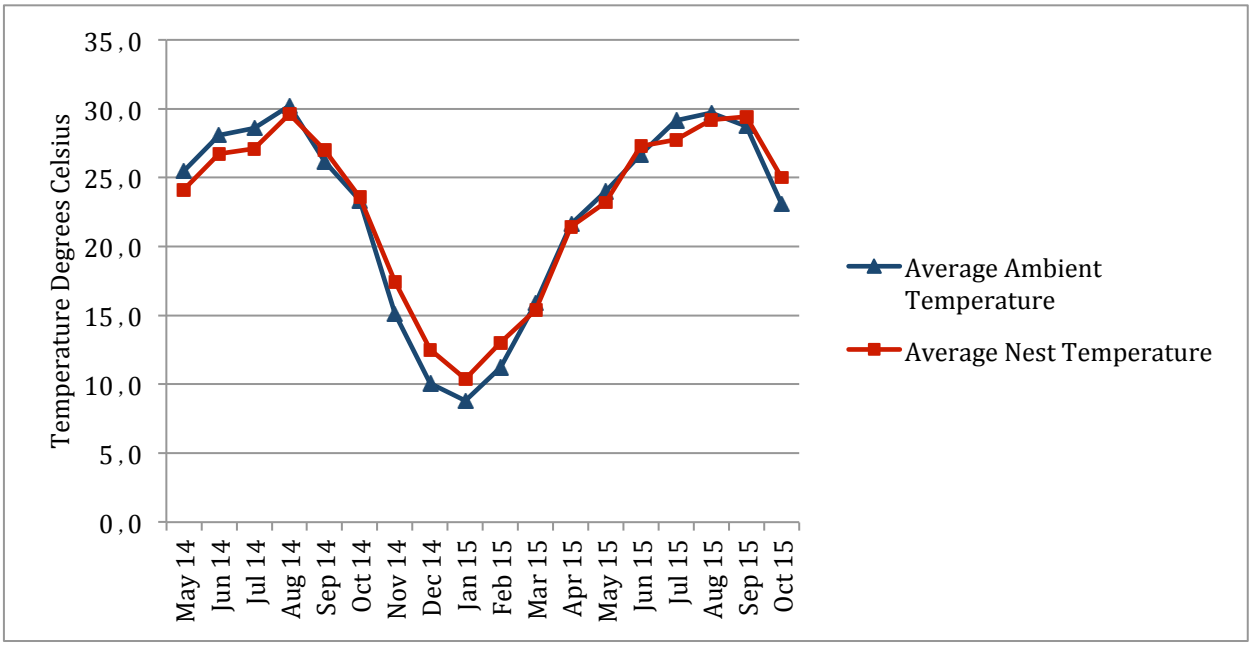


*Figure 1. Comparison of monthly average ambient air temperature with monthly average nest temperature, May 2014 – October 2015.*

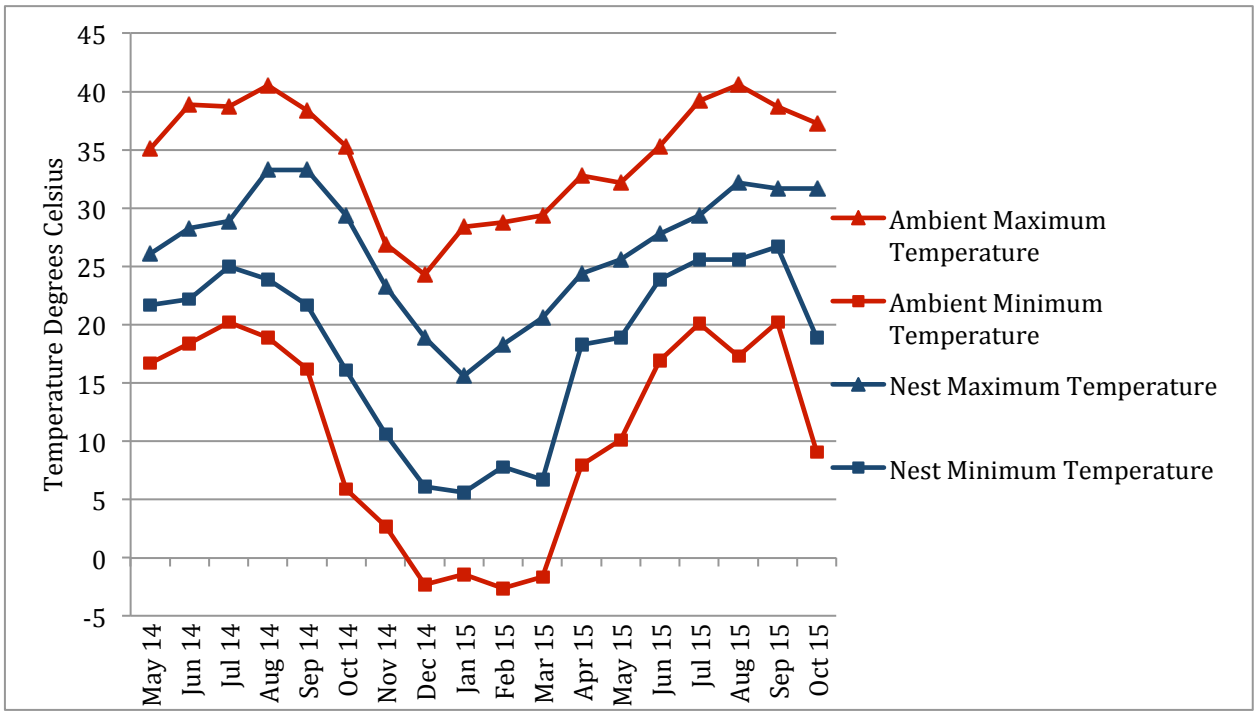


*Figure 2. Comparison of monthly maximum and minimum ambient temperature with monthly maximum and minimum nest temperature, May 2014 – October 2015*

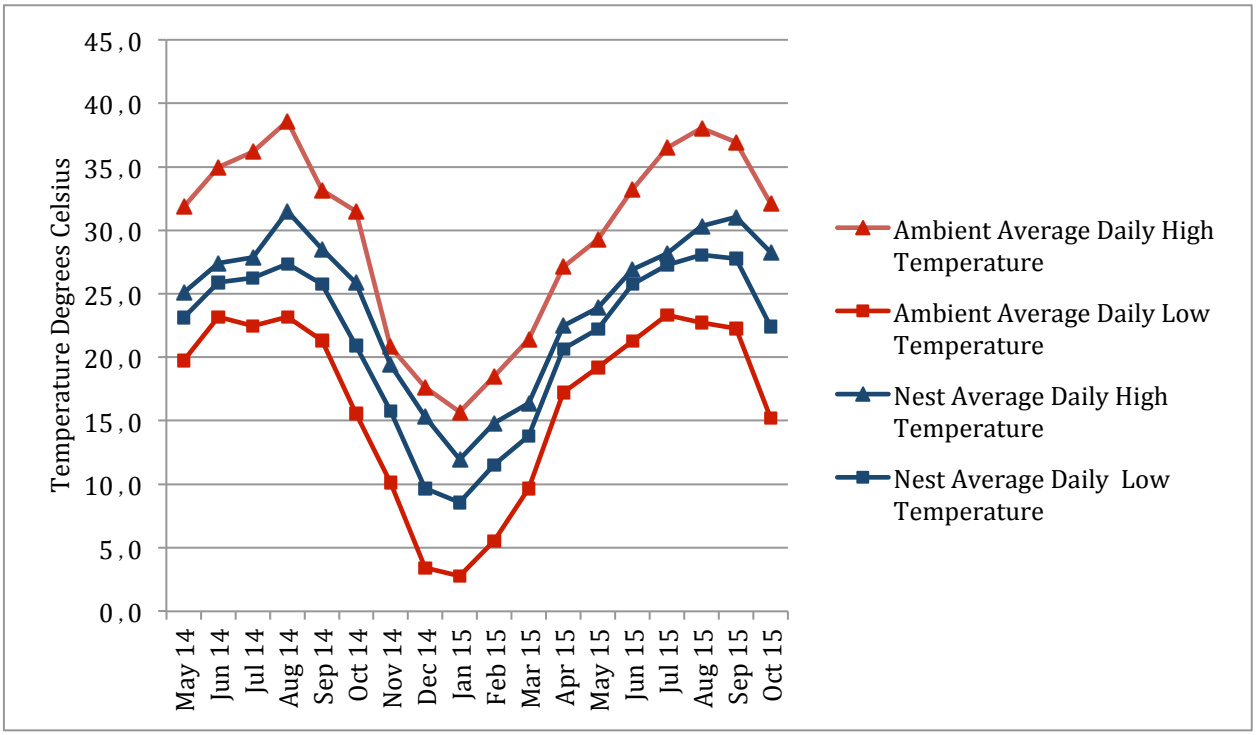


*Figure 3. Comparison of average daily high and low ambient temperature with average daily high and low nest temperature for each month, May 2014 – October 2015.*

*Correlation Analysis of Sandflies Captured per Trapnight and Temperature Data*

A Pearson's product movement correlation test was performed to determine if a significant relationship exists between sand flies captured per trapnight and ambient air temperature and woodrat nest temperature values. Concerning ambient temperature data, sand flies per trapnight strongly correlated with monthly maximum ambient temperature (Pearson's Correlation Coefficient = 0.658) and monthly average ambient daily high temperature (Pearson's Correlation Coefficient = 0.626) at the 0.01 level. Sand flies per trapnight strongly correlated with monthly average ambient temperature (Pearson's Correlation Coefficient = 0.547) at the 0.05 level. Sand flies per trapnight moderately correlated with monthly minimum ambient temperature (Pearson's Correlation Coefficient = 0.476) at the 0.05 level. Correlation between sand flies per trap night and monthly average ambient daily low temperature was not significant at the 0.05 levels. (Table 2)

Table 2
*Correlation of Sand Flies/Trapnight and Ambient Temperature Data*

| | Monthly Maximum Ambient Temp | Monthly Minimum Ambient Temp | Monthly Average Ambient Temp | Monthly Average Ambient Daily High Temp | Monthly Average Ambient Daily Low Temp |
|---|---|---|---|---|---|
| Sand Flies/Trapnight | | | | | |
| Pearson Correlation | .658** | .476* | .547* | .626** | 0.456 |
| Significance (2-tailed) | 0.003 | 0.046 | 0.019 | 0.005 | 0.057 |
| N | 18 | 18 | 18 | 18 | 18 |

*Note:* $*p < 0.05$, $**p < 0.01$.

Concerning woodrat nest temperature data, sand flies per trapnight strongly correlated with monthly maximum woodrat nest temperature (Pearson's Correlation Coefficient = 0.695) and monthly average woodrat nest daily high temperature (Pearson's Correlation Coefficient = 0.652) at the 0.01 level. Sand flies per trapnight strongly correlated with monthly average woodrat nest temperature (Pearson's Correlation Coefficient = 0.588), monthly minimum nest temperature (Pearson's Correlation Coefficient = 0.516) and monthly average woodrat nest daily low temperature (Pearson's Correlation Coefficient = 0.541) at the 0.05 levels. (Table 3)

Table 3
*Correlation of Sand Flies/Trapnight and Nest Temperature Data*

| | Monthly Maximum Nest Temp | Monthly Minimum Nest Temp | Monthly Average Nest Temp | Monthly Average Daily Nest High Temp | Monthly Average Daily Nest Low Temp |
|---|---|---|---|---|---|
| Sand Flies/Trapnight | | | | | |
| Pearson Correlation | .695** | .516* | .588* | .652** | .541* |
| Significance (2-tailed) | 0.001 | 0.028 | 0.010 | 0.003 | 0.020 |
| N | 18 | 18 | 18 | 18 | 18 |

*Note:* $*p < 0.05$, $**p < 0.01$.

# DISCUSSION

Increasing numbers of human Leishmaniasis cases are being reported in the USA every year. Leishmaniasis has been divided into three major forms: cutaneous, mucocutaneous, and visceral depending upon the species and the immune system of the host organism. Previous studies reported a number of native cases of cutaneous leishmaniasis in Texas (Clarke et al., 2013). Within the USA, leishmaniasis was first diagnosed in a 64-year-old woman in the late 1960s in Cameron County in the southern area in Texas. After that a number of domestic cases of cutaneous leishmaniasis have been reported in south central Texas since 1903. *L. mexicana* has been documented to be transmitted by sand flies of the *Lutzomyia* species. The mammalian reservoirs for disease transmission are woodrats of genus *Neotoma*. The history of all these cases revealed that none of the victims travelled outside of Texas, thus supporting the endemic aspect of the disease within Texas (McHugh, 1996).

When comparing our sand fly abundance results in this study to those of the previous study of McHugh, Ostrander, Raymond and Kerr, 2001, the total number of different species and the timing of the highest abundance of sand flies differs. In the study by McHugh et al. (2001) the sand fly species *Lutzomyia diabolica* and *Lutzomyia anthophora* were collected over periods of several months in Texas and analyzed in terms of their seasonal abundance. The sampling duration in Medina County was from April until October in the year 1997. The samples in Bexar County were collected over a year from April 1998 till December 1999. Of the two species studied, *Lu. diabolica* samples were obtained from April through November. During my study, I did not capture any individuals of *Lu. diabolica*. The highest abundance was witnessed in July. In the study by McHugh et al. (2001), *Lu. anthophora* samples were obtained from April through November, the highest abundance was witnessed in

September. This differs from my results in that I only captured *Lu. anthophora* between the months of July – October, with October being the month of greatest abundance. My results also differ from the results of McHugh et al. (2001) in that they captured individuals of *Lu. texana* over a span from April through October in 1997 with peak abundance occurring in April; whereas I only captured a total 2 *Lu. texana* and those were captured in August.

Further, strongest positive correlation with ambient air temperature with monthly maximum ambient temperature, monthly average ambient daily high temperature at the 0.01 levels ,and strongest positive correlation woodrat nest temperature with monthly maximum woodrat nest temperature, and monthly average woodrat nest daily high temperature at the 0.01 levels. This indicates that as temperature increases, sand fly abundance also is likely to increase.

The temperature environment inside of the woodrat nest was much more stable than the ambient air temperature. The temperature inside of the nest does not get nearly as hot or as cold on a daily basis as compared to the outside ambient temperature. This provides more stable environment for the sand flies, therefore, they may prefer to live down inside of the rat nest.

In this study investigate the temperature effects I found that increased sand fly abundance was associated with an increase in ambient air temperature and woodrat nest temperature. Sand fly distribution is limited in warm areas that have mean temperatures above 23°C. In Texas gradual increase in the temperature (Global warming) may increase sand fly abundance and activities, and may have profound effects on sand fly populations. Therefore, small changes in temperature can expand the geographical area in which the *Leishmania* is able to replicate, allowing expansion of endemic areas.

# REFERENCES

Alvar, J., I. D.Vélez, C . Bern, M . Herrero, P . Desjeux, J . Cano. (2012).WHO *Leishmaniasis* Control Team. Leishmaniasis worldwide and global estimates of its incidence. *PloS One*, **7**(5), e35671.

Chappuis. F., S. Sundar, A. Hailu, H . Ghalib, S. Rijal, R . W. Peeling, J. Alvar, and M. Boelaert., 2007.Visceral leishmaniasis: what are the needs for diagnosis, treatment and control? Microbiology. 5: 873–882.

Clarke, C. F., K. K. Bradley, J. H . Wright, and,J . Glowicz. (2013). Case report: Emergence of autochthonous cutaneous leishmaniasis in northeastern Texas and southeastern Oklahoma. The American Journal of Tropical Medicine and Hygiene, 88: 157–161.

Desjeux, P.1996. Leishmaniasis. Public health aspects and control. 14: 417–423.

Dostálová, A., and P.Volf., 2012. Leishmania development in sand flies: parasite-vector interactions overview, Dostálová and Volf Parasites & Vectors 5: 276.

Endris, R. G., D . G .Young,and, P. V. Perkins, 1987. Experimental transmission of *Leishmania mexicana* by a North American sand fly*, Lutzomyia anthophora* (Diptera: Psychodidae). Journal of Medical Entomology, 24: 243–247.

González C, O . Wang, S . E Strutz, C . González-Salazar , V . Sánchez-Cordero , et al. 2010. Climate Change and Risk of Leishmaniasis in North America: Predictions from Ecological Niche Models of Vector and Reservoir Species. PLoS Neglected Tropical Diseases 4: e585.

González, C., E. A. Rebollar-Téllez, S . Ibáñez-Bernal, I . Becker-Fauser,E. Martínez-Meyer, A. T. Peterson, and V.Sánchez-Cordero, 2011. Current Knowledge of *Leishmania* Vectors in Mexico: How Geographic Distributions of Species Relate to Transmission Areas. The American Journal of Tropical Medicine and Hygiene, 88: 839–846.

Gratz, N. G. 2006. Vector- and Rodent-Borne Diseases in Europe and North America: Distribution, Public Health Burden, and Control. Cambridge University Press.

Handman, E. 2001. Leishmaniasis: current status of vaccine development. Clinical Microbiology Reviews. 14: 229–243.

Hernández-Torres, Alicia, E . García-Vázquez, J . Bravo-Urbieta, E. B. Morell, B . Alcaraz-Vidal, A . Sánchez-Serrano, and J . G . Gómez. 2015. "La leishmaniasis visceral en la región de Murcia: estudio multicéntrico 1997-2013." 19 :24-30.

Jeziorski, E., J. Dereure, G. Mac Bullen, C. Blanchet, C. Ludwig, V. Costes, & M. Rodière. 2015. "Mucosal relapse of visceral leishmaniasis in a child treated with anti-TNFα." International Journal of Infectious Diseases 33 :135-136.

Kerr, S. F., C. P. McHugh, & R. Merkelz.1999. Short report: a focus of *Leishmania mexicana* near Tucson, Arizona. The American Journal of Tropical Medicine and Hygiene, 61: 378–379.

Kumari, S, F. Jamal, P . Shivam, A. Thakur, M. Kumar, S. Bimal, V. N. R. Das, K. Pandey, S. Narayan, A. K. Gupta, P. Das, & S. K. Singh. 2015. *"Leishmania donovani* skews the CD56+ Natural Killer T cell response during human visceral leishmaniasis. Cytokine 73:53-60.

Lawyer, P. G., D. G . Young, J. F.Butler, & D. E.Akin, 1987. Development of *Leishmania mexicana* in *Lutzomyia diabolica* and *Lutzomyia shannoni* (Diptera: Psychodidae). Journal of Medical Entomology, 24: 347–355.

McHugh, C. P. 2010. Cutaneous leishmaniasis in Texas. Journal of the American Academy of Dermatology, 62: 508–510.

McHugh, C. P., B. F. Ostrander, R. W. Raymond, & S. F. Kerr, 2001. Population dynamics of sand flies (diptera: psychodidae) at two foci of leishmaniasis in Texas. Journal of Medical Entomology, 38: 268–277.

McHugh, C. P., P .C. Melby, & S . G . LaFon. 1996. Leishmaniasis in Texas: epidemiology and clinical aspects of human cases. The American Journal of Tropical Medicine and Hygiene, 55: 547–555.

Neghina, R., & M. N. Neghina., 2010 Leishmaniasis,a global concern for travel medicine,42: 563–570.

Nzelu, C. O., A. G. Cáceres, M. J. Arrunátegui-Jiménez, M. F. Lañas-Rosas, H. H. Yañez-Trujillano, D. V. Luna-Caipo, C. E. Holguín-Mauricci, K. Katakura, Y. Hashiguchi, and H. Kato.2015. "DNA barcoding for identification of sand fly species (Diptera: Psychodidae) from leishmaniasis-endemic areas of Peru." Acta Tropica 145:45-51

Raymond, R. W., C. P. McHugh, L. R.Witt, and S. F.Kerr. 2003. Temporal and spatial distribution of *Leishmania mexicana* infections in a population of *Neotoma micropus*. Memórias Do Instituto Oswaldo Cruz, 98:171–180.

Roberts L. S. and Janovy, J. Jr. 2009. Gerald D. Schmidt & Larry S. Roberts' Foundations of Parasitology, 8th ed. New York (NY): McGraw-Hill

Ryan, E. T., T.Solomon, and D. R. Hill, 2012. Hunter's Tropical Medicine and Emerging Infectious Disease,Expert Consult - Online and Print,9: Hunter's Tropical Medicine and Emerging Infectious Disease. Elsevier Health Sciences.

United States. Bureau of Soils. Soil map, Texas, Wilson County, Map, June 9, 1908

Wright, N. A., L. E. Davis, K.S. Aftergut, C.A. Parrish, and C.J. Cockerell. 2008. Cutaneous leishmaniasis in Texas: A northern spread of endemic areas. *Journal of the American Academy of Dermatology*, 58:650–652.

Young, D. G., and M. A. Duncan. 1994. Guide to the identification and geographc distribution of Lutzomyia sand flies in Mexico, the West Indies, Central and South America (Diptera: Psychodidae). Associated Publishers, Gainesville, Florida, USA. 881p.