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Research Question:
Currently I have two research questions I am working on.
1. What is the current level of tickborne disease in the ticks on the TAMIU campus, and other locations in Webb County?
2. What are the correlations between tick activity and environmental variables.

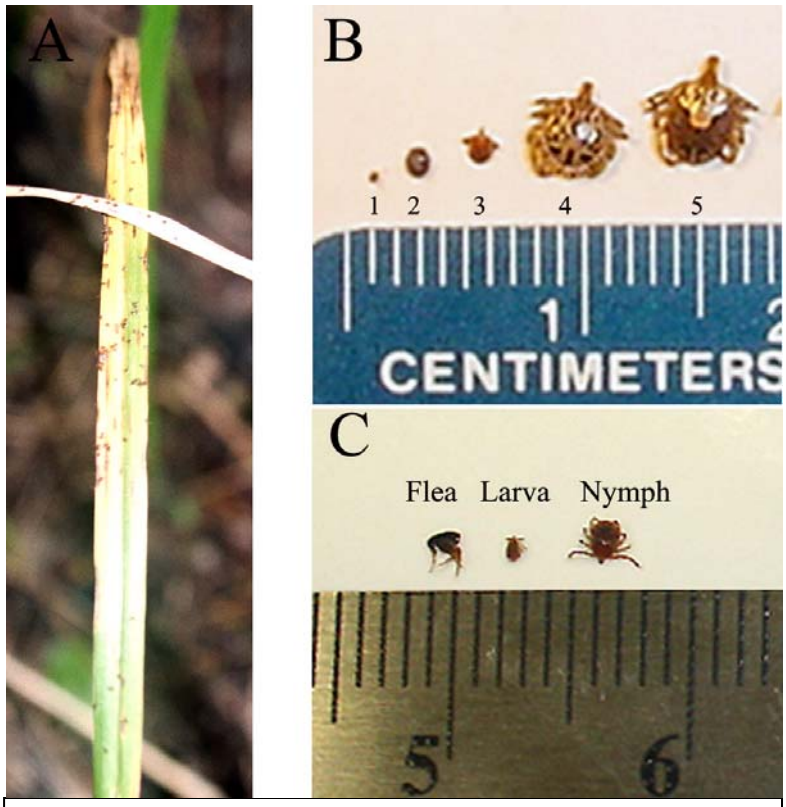

Figure 1.
A. There are over 60 ticks on this blade of grass. (*A. cajennense* larval ticks questing on a blade of grass.)
B. *A. cajennense* ticks life stages: 1. larval tick, 2. fully engorged larval tick, 3. nymph, 4. adult male, 5. adult female.
C. A flea, larval tick and nymph shown for size comparison. Scale in centimeters.

What are ticks? Ticks are blood-sucking arachnids commonly found in wooded and grassy areas throughout the United States. Ticks are a major vector of infectious disease, including ehrlichiosis and Rocky Mountain Spotted Fever. There are two major kinds of ticks, ticks with a hard body called hard ticks (Ixodid Ticks) or ticks with a soft body called soft ticks (Non-Ixodid Ticks) (1). Ticks hatch from an egg as a larva, grow into a nymph, and finally mature into an adult. At each stage of the hard tick's life they take a single blood meal. Often during taking the blood meal they acquire a pathogenic organism which they transmit, in their subsequent blood meal to another host. Additionally, some tick-borne diseases are transmitted from the adult female to the eggs that she lays. This means that the newly hatched larva is already carrying and will transmit the pathogen. The larvae are particularly difficult to notice on a human host since they are only slightly larger than the period at that end of this sentence (Figure 1).

Importance: Ticks are the second most common vector of infectious diseases. Ticks feed on the host for 3-7 days causing little to no itching or pain. Often the tick bites often go unnoticed for several days. During this time they may transmit tickborne diseases to the host. The long term goal of this project is to complete an analysis of the disease prevalence in ticks isolated from the TAMIU campus and other locations in South Texas. We have field sites currently in Jim Hogg, Maverick and Webb County.

What is the current level of tick activity on the TAMIU campus?

The PI and students collected and speciated 37,236 free-living ticks from March 2005 to October 2007. *A. cajennense* represented over 98.7% of the ticks collected from Webb County, Texas (Table 1). The primary means of collection is through the use of carbon dioxide traps on the TAMIU campus. Out of 37,236 ticks speciated 36,765 were *A. cajennense* and only 1 was an *A. americanum* adult.

Table 1: Ticks from TAMIU 2005-2006

Tick Species	# Collected	% of Total
Amblyomma cajennense	22459	98.3%
Amblyomma inornatum	236	1.0%
Amblomma maculatum	3	0.01%
Dermacentor albipictus	45	0.2%
Dermacentor variabalis	57	0.2%
Haemaphysalis leporispalustris	33	0.1%

Table 2: Ticks from other sites

County	Webb				Maverick	Jim Hogg
Tick Species	**Site 1**	**Site 2**	**Site 3**	**Site 4**		
Amblyomma cajennense	510	2	6	107	6	200
Amblyomma inornatum	0	0	0	4	13	3
Dermacentor albipictus	7	0	0	0	0	0
Dermacentor variabalis	4	0	1	1	5	0
Haemaphysalis leporispalustris	0	0	0	1	0	1

Preliminary data from other field sites in Webb County (Table 2) show similar trends. Preliminary data from Jim Hogg County is similar to Webb County, but the data from Maverick County initially shows less *A. cajennense* than other species. The data from these field sites is insufficient at this time to draw conclusions. Data from additional field sites in this region would be needed to determine the distribution of *A. cajennense*.

The vast majority of those ticks collected (98.3 %) were *Amblyomma cajennense* (Table 1). *Amblyomma cajennense* (the Cayenne Tick) is of serious concern, to quote one researcher, “all stages feed aggressively on people” (17).

Field Observation: Field observations of *A. cajennense*.

A. cajennense has been previously described as having a clear association with grass height and tick abundance. There was a lack of association in forest type areas with heavy brush reported in Trinidad (3). In contrast, in Brazil an increased abundance of *A. cajennense* in forested areas was seen compared to open areas (4). Preliminary results

confirm the association with grass height in Webb County. No correlation of tick activity was seen with native grasses, but significant correlation of activity was seen with the presence of Buffelgrass. In South Texas Buffelgrass routinely grows to over 1 meter in height. When considering brush density, little tick activity was observed in open fields, with abundant tick activity observed in river beds forest/dense vegetation or in surrounding fields. It is likely that the tropical nature of the Trinidad field sites decreased the need for forested areas. In Webb County, nymph and adult ticks are found in a dispersed pattern, whereas larval ticks are found in clusters, usually in more shaded areas. This is consistent with observations in other species of ticks (5, 6, 7). When larvae are found through tick walks (literally on the researcher's clothes while the researcher is walking through the study site) the larval ticks are typically found to be questing 20-30 cm above ground level. Figure 1A shows the larval *A. cajennense* ticks on a blade of Buffelgrass approximately 30 cm long. This is consistent with *A. cajennense* preferentially feeding on medium to large sized mammals at all life stages (2, 4, 8, 9, 10).

Weather Observations:
The PI has completed a two-year study of the climatic variables and their correlation with tick activity. The first 18 months of this five year study were presented by a student working in the lab at a national meeting (11).

As can be seen in figure 2 (Top) there was a peak in larval activity during the fall of year one, and during the late-spring of year two. This peak correlates with rain events ($p<0.004$). The first year is consistent with previous observations that larva hatch during the fall in Brazil (12), the second year is not. Interestingly, in looking at the rain data in that paper, larval activity was seen after several weeks of no rain during the fall (12). However, this

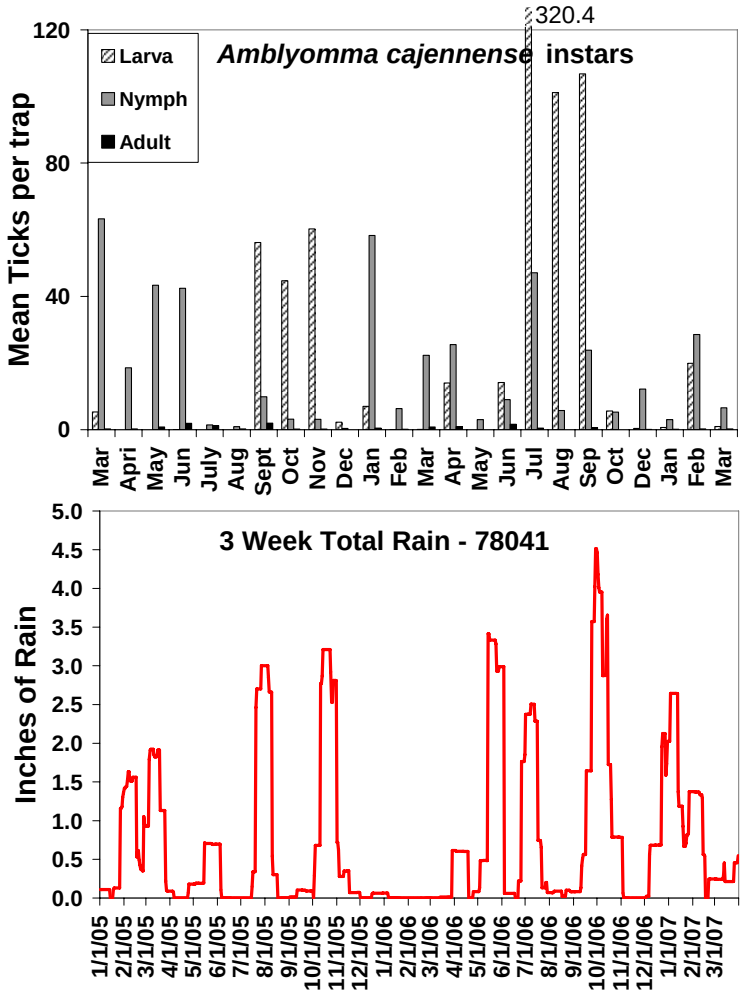

Figure 2 Correlation of tick activity with weather data. The amount of rain during the prior 3 week period was positively correlated ($p<0.004$) with larva activity.

area of Brazil receives over an inch of rain per week, and Webb County receives about an inch or less on average per month, with long periods of drought being common (see Figure 3 Bottom). Note how the graph shows sudden increases in the three week average corresponding with 0.5-6.0 inch rain events. Large rain events were observed in year one (2005) in August and October, with a corresponding larval peak in September-November. In year two, rain in April, June and July led to a larval peak in the summer (July-August). In the beginning of year three rain events in January and February correspond to a peak in February. A five month drought was observed from November 2005 to April 2006. Droughts lasting 2-3 months are common. It appears that the lack of rain prevents emergence of the larva, but it is interesting to hypothesize that the presence of rain also inhibits the emergence of the larva. The larval ticks seem to emerge after the cessation of the "rainy season." More data will be needed to gain a better

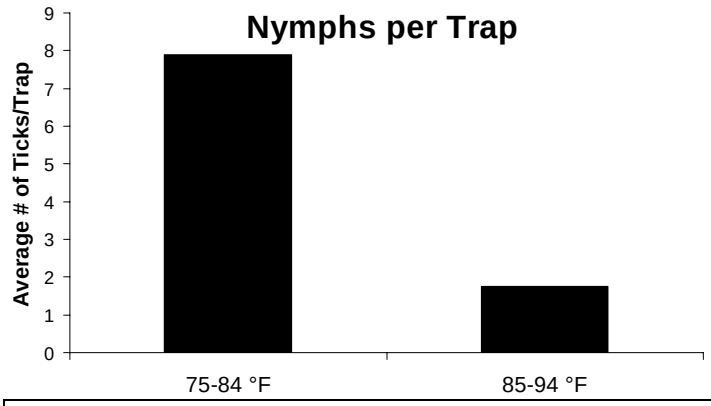

Figure 3 – Correlation of tick activity with weather data. The mean nymphs per trap and temperature. Not significant.

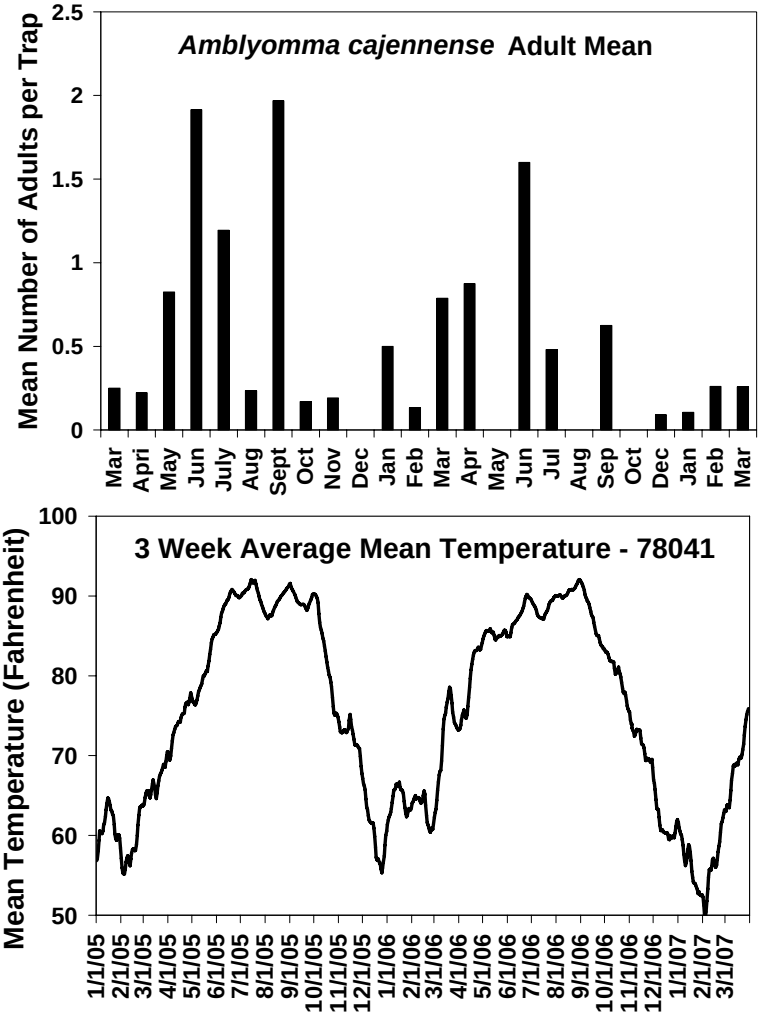

Figure 4 – Correlation of tick activity with weather data. The 3 week average mean temperature was positively correlated with adult tick activity.

understanding of the variables involved in larval tick emergence. How much rain is needed, and how much rain is inhibitory for emergence.

The activity of nymph *A. cajennense* showed an interesting correlation with temperature. Nymphs were more commonly seen at moderate temperatures than at very hot or very cold temperatures (Figure 3). This correlation did not reach significance. This may reflect the decreased survival advantage of nymphs at hot dehydrating temperatures as compared to adults which are positively correlated with increased temperature.

As seen in Figure 4, adult *A. cajennense* shows a positive correlation with increased temperature (3-week mean $p<0.014$). The adults are more abundant in the summer months. However, they are present throughout the year. This combined with the fact that larval ticks emerge at different times throughout the year suggest that it may be possible that *A. cajennense* is completing more than one life cycle per year (discussed below). Not surprisingly, overall tick activity was positively correlated with wind ($p<0.003$) and is likely due to more effective dispersion of the carbon dioxide from the trap.

A. cajennense is capable of completing its life cycle twice per year in the laboratory (13). Field observations in Brazil and Argentine indicated that it had a single cycle per year with a clear diapause (4, 12). It is clear that at least one life cycle is completed per year in South Texas, but it is possible that more than one cycle may be completed per year, or that there is more than one overlapping cycle occurring simultaneously. Given the irregular nature of the pattern observed during this two year period of time it will be necessary to look at tick activity in this region over a longer period of time to determine the natural cycle of this tick in the South Texas Plain and Tamaulipan Biotic Province. No clear diapause is apparent at this time. Additional data is needed to get a clear understanding of the natural life cycle of *A. cajennense* ticks under semi-arid conditions. The data currently available from Brazil and Argentina is not generalizable to South Texas.

Animal Data

A preliminary survey of domestic animals, and local wildlife was conducted from 2005-2007 in collaboration with Dr. Vaughan and his mammalogy students. A total of 54 mammals were inspected for ectoparasites (methods discussed below). Mammal species inspected: black-tailed jackrabbit (*Lepus californicus*), Mexican ground squirrel (*Spermophilus mexicanus*), raccoon (*Procyon lotor*), cotton tail rabbit (*Sylvilagus audubonii*), house mouse (*Mus musculus*), cotton rat (*Sigmodon hispidus*), opossum (*Didelphis virginiana*), pocket mouse (*Chaetodipus hispidus*), peccary (*Tayassu tajacu*), bobcat (*Lynx rufus*), dog (*Canis lupus familiaris*), house cat (*Felis silvestris catus*), and Texas longhorn (*Bos Taurus*). Animals were from Webb County and Zapata County (immediately to the south of Webb County). This initial survey identified *A. cajennense* adults on the peccary (9) and dog (1 plus 2 nymphs). This is consistent with previous reports (13, 14, 15).

Table 3: Ticks Collected from Mammals in South Texas (Webb and Zapata Counties)								
Host (Number)	*Amblyomma cajennense*	*Amblyomma inornatum*	*Amblyomma maculatum*	*Argasidae*	*Dermacentor variabilis*	*Ixodes*	*Haemaphysalis leporispalustris*	*Rhipicephalus sanguineus*
House mouse (3)	0	0	0	0	0	0	0	0
Domestic cat (2)	0	0	0	1	7	0	0	0
Domestic dog (22)	3	0	0	0	3	0	0	232
Cotton Rat (4)	0	0	0	0	0	0	0	0
Pocket Mouse (2)	0	0	0	0	0	0	0	0
Mexican Ground Squirrel (4)	0	0	0	0	0	0	3	0
Opossum (3)	0	0	0	0	1	0	0	0
Racoon (2)	0	18	0	0	106	0	5	0
Cotton Tail Rabbit (4)	0	4	0	0	0	0	3	0
Jackrabbit (4)	0	0	0	0	0	17	23	0
Bobcat (2)	0	14	0	0	9	0	0	0
Peccary (1)	3	0	0	0	10	0	0	0
Longhorn (1)	0	0	1	0	0	0	0	0

PCR Data. Initially 102 *Dermacentor variabilis* were screened in the laboratory of Dr. Philip Williamson so that the PI could learn the methods for tick DNA purification and PCR protocols. Three ticks were positive: 1/102 was positive for *Borrelia lonestari*; 3/102 were positive for *Rickettsia rhipicephali*; none were positive for *Ehrlichia* or Lyme Disease. Positive samples were confirmed by sequence analysis.

Subsequently, the PI and students have screened 165 pools (representing 534 ticks) of *A. cajennense* for *Borrelia lonestari* with one positive pool; 166 pools (representing 547 ticks) of *A. cajennense* for Spotted Fever Group Rickettsia with 6 positive pools (all pools represent 1-8 ticks, sequence confirmation is pending Table 4). Currently the vast majority of the ticks examined are from the primary field site, TAMIU campus in Webb County. The level of Spotted Fever Group Rickettsia is less than 10% and *B. lonestari* at this field site is less than 2%. It is not uncommon to find one field site with a low level of prevalence and a second field site with a high level of prevalence (16). For this reason analysis of multiple field sites is ongoing (preliminary results are in Table 4).

Table 4. Spotted Fever Group Rickettsia					
County	**Webb**			**Jim Higg**	**Total**
	Site 1	**Site 2**	**Site 3**	**Site 1**	**All Sites**
Adults (n=1)	**0 / 4**	**0 / 78**			**0 / 82**
Nymphs (n=5)	**0 / 15**	**6 / 35**	**0 / 5**	**0 / 14**	**6 / 69**
Nymphs (n=8)		**0 / 15**			**0 / 15**
Prevalence	**0-2%**	**1-10%**	**0-4%**	**0-2 %**	

Number positive / Number Screened

Regional Significance: A recent seroprevalence study conducted in Coahuila, Nuevo Leon and Tampaulipas, Mexico demonstrated a 6%, 3.9%, and 14.1% with antibodies against *Borrelia burgdorferi* by western blot. *Borrelia burgdorferi* is the causative agent of Lyme disease (19). This is a very high prevalence rate for a single tick borne disease considering that Lyme disease is rarely diagnosed in Texas with only 639 cases diagnosed between 1986 and 1996 in the entire state (18). This sharp contrast between the number of cases diagnosed in Texas and the high seroprevalence rate across the border from South Texas demands a careful evaluation of tickborne diseases in South Texas. If we do not know what is going on in South Texas how do we know what our risk is.

Our preliminary results show that there is a low prevalence of *Borrelia lonestari*, *Rickettsia rhipicephali* and potentially other Spotted Fever Group Rickettsia in Webb County. In late 2007 a small outbreak of ehrlichiosis was reported in Webb and Zapata counties. We are in the process of obtaining the control samples to begin screening for this disease.

How can we protect ourselves from ticks?

Ticks are present in the environment Figure 5. We like to enter that environment for recreational and work purposes. General recommendations can be made:

Protect yourself and control ticks. These five recommendations are from the Texas Cooperative Extension (20):

Figure 5: This picture shows one of the preferred habitats of the tick. The leaves here are about the size of the last digit of your fingers. There is abundant leaf litter, although it is not very deep as the ground can be seen below the litter.

Recommendations:

1. Make it harder for ticks to enter clothing by tucking shirttails inside pants and wearing long pants and shirts.
2. Use duct tape or wide masking tape to seal pants cuffs to boots. Tucking your pants legs inside your socks is also effective.
3. Avoid sitting on the ground or on logs in brushy, tick-infested areas. Ticks often crawl around on a host for hours before biting. When in tick-infested areas, have a friend check you frequently for ticks before they attach.
4. Around the home, keep tall grass and weeds cut short. Ticks like to climb vertical surfaces to rest after feeding. When treating yards for brown dog ticks, spray the siding of the house, fences, trees and other hiding places as well as the lawn.

5. When using insecticides, follow label directions carefully and do not apply more than is recommended. Tick insecticides can be dangerous to pets and children if misapplied.

What should we do if we find a tick on our body?

Of concern is the response of the public to ticks found on their body. Tick borne diseases can be transmitted quickly, although most transmit slowly. Thus prompt removal of the tick will avoid most disease transmission. This is often not done. It is common to hear of people crushing ticks, smashing them, etc. This practice is actually very unsafe as it releases the infectious agent onto the persons hands, or environment. This will allow for potential disease transmission.

Best Approach: Gently remove the tick from the skin using a pair of forceps. Do not squeeze the body of the tick, but grab it right where it is attached to the skin. It may take many gentle tugs to successfully remove the tick. The tick can then be placed in any convenient sealable container like a Zip-Loc bag. Do not crush the tick. Importantly, wash your hands as they sometimes become contaminated with blood during the removal process. The tick bite should be reported to your doctor and the tick sent to the state lab for pathogen detection.

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