# Session 7

# The Black Cherry Tree/Eastern Tent Caterpillar/ Eastern Tent Caterpillar Frass System

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# The Biology of the Tent Caterpillar As It Relates to Mare Reproductive Loss Syndrome

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DURING THE SPRING OF 2001, HORSE FARMS IN CENTRAL Kentucky experienced an unprecedented loss of foals. Affected horses exhibited signs collectively referred to as Mare Reproductive Loss Syndrome (MRLS). Concurrent with the onset of MRLS was a population explosion of the eastern tent caterpillar (ETC), which drove starving caterpillars from the canopies of defoliated trees into adjacent pastures housing pregnant mares. Although the exact cause of MRLS remains unknown at this time, there is a growing consensus based on epidemiological surveys and preliminary experimentation that the caterpillar may have played a central role in the outbreak of MRLS. It is the purpose of this paper to outline the basic features of the biology and population dynamics of ETC as they relate to MRLS and to consider specific ways in which the caterpillar might cause harm to vertebrates. Unless otherwise referenced, the information presented here is derived from (1) and references therein.

Twenty-six species of tent caterpillars (*Malacosoma:* Lasiocampidae) occur in the northern latitudes of both the New and Old World. Six species are found in North America, but the only species to occur in Kentucky are the forest and ETC. The forest tent caterpillar (*M. disstria*) has the largest range of all the North American species, while the ETC (*M. americanum*) is largely limited to the eastern half of the United States and Canada. The two species are readily distinguished by larval color patterns, host choice, and behavior. *M. disstria* is the only species of tent caterpillar that does not construct a communal silk tent; sibling aggregates rest gregariously in the open on leaves and on the bark of the host tree.

Local population of forest and ETC are found wherever their host species occur. The ETC is largely restricted to trees in the plant family Roseaceae, greatly preferring the black cherry but may also oviposit on choke cherry, fire cherry, apple, plum, peach, and pear, and more rarely on hawthorn, flowering quince, mountain ash, and *Cotoneaster*. Starved caterpillars and full-grown caterpillars that have dispersed from their natal tree may attempt to feed on other species of plants as well. The forest tent caterpillar may also feed on fruit trees, including black cherry, but prefers tupelo in the Southeast, sugar maple in the Northeast, and poplar in the North Central states and Canada. Because of its broad host range, which includes many important species of forest trees, the forest tent caterpillar is considered the most economically important of the North American tent caterpillars. However, it has thus far not been implicated in any naturally occurring instance of MRLS.

All species of tent caterpillars have similar life cycles (Figure 1). The overwintered eggs of the insect hatch in the early spring, just as the buds break and leaves begin to unfold, but it is not unusual for the caterpillars to emerge even before leaf flush and to subsist by mining the buds of the plant. The caterpillars are adapted to the chemistry of young leaves, placing a premium on early emergence.

ETC build a communal silk shelter (tent) that they expand daily to accommodate their growth. The caterpillars are central place foragers, and all activity is centered about the tent. When young, colonies launch forays to feeding sites distant from the tent in the afternoon, at dusk, and in the early morning before dawn. In the last instar, caterpillars from undisturbed colonies typically feed only at night, returning to their tent at dawn. The caterpillars are highly social and conduct all their activities in tight synchrony, moving to food, feeding, and returning to the tent en masse. After feeding, they rest together in or on the tent until their next foraging bout.

The caterpillars are ravenous feeders, completing their larval growth in as few as seven to eight weeks. Indeed, the ETC is among the fastest growing of all caterpillars. In the field, growth is constrained by low seasonal tempera-



**Figure 1.** Seasonal life history of the ETC in Kentucky. The exact time of egg hatching is temperature dependent and varies from year to year.

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tures, but in the laboratory at room temperature, the larvae reach their full size in only three weeks, increasing in mass during this period by a factor exceeding 5,000. When fully grown, the caterpillars drop from their natal tree, often traveling long distances over the ground in search of sheltered sites. Here the caterpillars spin cocoons and metamorphose, spending two to three weeks in the pupal stage before emerging as moths.

The adult moth is short lived and does not feed. The female emerges from the cocoon in late afternoon and secretes a sex pheromone to call males. Impregnated females typically oviposit that same evening. Thus, the female may emerge, mate, and oviposit in rapid succession, and being completely spent, die shortly thereafter.

The moth produces a single egg mass that contains approximately 300 eggs, but the exact number is highly variable. The eggs are wrapped around a branch of the tree near its tip and covered with spumaline, a frothy secretion from the female's accessory gland. The material serves to protect the eggs from parasitic wasps and desiccation.

Although embryogenesis occurs soon after the eggs are laid in June or July, the small larvae lie quiescent until the following spring. Thus, the most durable life stage of the insect is the small, inactive caterpillar that spends nearly nine months sequestered within the shell of its egg. For those concerned with managing populations of the insect, the caterpillar is most vulnerable during this stage of its life. The egg masses are readily seen after the leaves have fallen from trees in the autumn and can easily be cut from the branches and discarded. Indeed, this was a favored method of control in the early part of the twentieth century when entomologists approached the control of the caterpillar with a near missionary zeal. In some areas, contests were held and school children rewarded for every egg mass they collected.

Wherever they occur, ETC have boom-and-bust population dynamics; outbreaks alternate with periods of scarcity. Because of its short life cycle and the capacity of a single female to produce a large number of eggs, the reproductive potential of the ETC is enormous. In theory, a single pair of moths can produce a population of nearly 100 million caterpillars in only four seasons. Although local populations never fully achieve their full potential for exponential growth, they typically reach high densities for several years in succession, then collapse and all but disappear. Records collected for well over a century show that the duration of outbreaks and the interval between them is highly variable. The most complete records, complied for the forest tent caterpillar, indicate that outbreak populations may last for as little as one year to as many as nine.

The irruptive population dynamics of tent caterpillars are attributable to both density-dependent and densityindependent mortality factors. The major density-dependent mortality factors are predation, disease, and starvation. The ETC has more than 200 known predators and parasitoids. They are often present in abundance during the late stages of outbreaks, and some have been credited with bringing population surges to an abrupt end. The caterpillar is also susceptible to viruses, bacteria, and microsporidians, particularly at high population densities.

When population densities peak, ETC commonly defoliate their natal trees well before they are fully grown, forcing them to strike off over the ground in search of a new food plant, an endeavor in which they are rarely successful. The caterpillars show great initial reluctance to leave their existing trail system and typically make little progress through the physically complex ground cover. Even after several days, colonies may succeed in traveling only a few meters from the natal tree. During these offthe-tree forays, the caterpillars initially maintain their social instinct, moving only short distances from their siblings and marking their progress with silk and pheromone pathways that others follow. The caterpillars have rudimentary eyes and can visualize the silhouettes of plant stems from short distances, and they may attempt to orient visually to them. It is likely but untested that when moving through pastures, the caterpillar may mistake the legs of horses for tree stems and orient to them as well. Regardless of their efforts, dispersing caterpillars rarely find another suitable host. They typically become isolated from their siblings and marooned on unpalatable plants where they eventually starve or are killed by predators.

Inclement weather is the preeminent density-independent mortality factor affecting populations of tent caterpillars. This is the case because the caterpillars cannot process food when their body temperatures fall below 15°C. To achieve the body temperatures needed to digest food, the caterpillars bask in the sun on cold spring days, either on or in their tent. Their black bodies absorb solar radiation, and the protective layers of the tent minimize convective heat loss. If the days are cold and cloudy, the caterpillars grow little or not at all, and if these conditions persist for two weeks or more, colonies begin to perish. Occasionally, late-season frost kills the leaves of the host tree, and the caterpillar populations, left without food, experience a region-wide die-off. Many instances of population collapses due to cold springs have been documented in the northern part of the ranges of both the forest and ETC.

When a tentative association between the ETC and MRLS was first made in 2001, the caterpillars were near the peak of their population cycle in many of the affected areas. Reports from the field indicated that pastures were overrun with dispersing caterpillars. Masses of caterpillars were observed walking along the rails of fences, and many fell into troughs used to water horses. Thus, it became of urgent interest to explore all aspects of the biology of the caterpillar in an attempt to determine what attribute of the insect might link it to MRLS (Table 1). One of the first hypotheses to be to tested was the possibility that the caterpillars had poisoned pregnant mares with cyanide, triggering abortions and stillbirths. The principal host of the ETC, the black cherry, is capable of producing cyanide in response to herbivory. The leaves of the tree contain the cyanogenic glucoside, prunasin. When ingested by ETC, or any other herbivore, enzymes in the leaf convert prunasin to mandelonitrile, then to benzaldehyde and cyanide. A recent study showed that at the time of the year when ETC feed on cherry trees, the leaves have an average cyanide potential (HCN-p) of 1,900 ppm (2). Young leaves at the tips of branches have a mean HCN-p of more than 3,000 ppm, while oldest leaves at base of the stems have an HCN-p of about 1,100 ppm. ETC prefer the youngest and most cyanogenic leaves. When allowed to feed overnight on young leaves, the bolus of the foregut contained an average about 600 ppm cyanide the next morning, while the midgut bolus contained an average of only 14 ppm (2). The caterpillars are unable to digest much of the leaf, and approximately 50% of the energy of the leaf is egested as fecal pellets. When dry, the pellets contain less than 100 ppm of cyanide. Thus, the ETC quickly detoxifies cyanide, and compared to the amount of cyanide that an herbivore might ingest if it fed on cherry leaves, it would acquire relatively little cyanide if it inadvertently consumed ETC or their fecal pellets. Most damaging to the cyanide hypothesis was the result of another study showing that when pregnant mares were treated with higher concentrations of cyanide than they could reasonably be expected to assimilate by ingesting caterpillars or their fecal pellets, they exhibited no signs of MRLS (3).

Another attribute of ETC having the potential to cause harm to animals that come in contact with them is its hairs or "setae." Some caterpillars, such as those of the buck moth (Hemileuca maia), have poisonous hairs that sting the victim when touched. These "urticating" hairs act like hypodermic needles, bearing a sharp point that penetrates the skin and a hollow shaft filled with poison. The most notorious of the urticating caterpillars is the pine processionary of southern Europe (Thaumetopoea pityocampa), whose larvae form long, head-to-tail processions as they move over the ground in search of pupation sites. Compared to the buckmoth, the hairs of the pine processionary are lighter and more brittle, scatter easily in the wind, and can affect individuals who have no direct contact with the caterpillars. It was recently discovered that susceptible individuals are also capable of exhibiting an immunologic response to a setal protein carried by the processionary, and instances of anaphylaxis have been reported (4). In contrast to the setae of the caterpillars of the buckmoth and pine processionary,

Table 1. Attributes of ETC potentially capable of harr	ning
vertebrates.	

-	Pat	hway				
Attribute	Ingestion	Inhalation	Mode of Action			
cyanide	<b>x</b> <sup>1</sup>		toxin			
calcium oxalate	<b>x</b> <sup>1</sup>		toxin			
cuticular ketosteroids	<b>x</b> <sup>1</sup>		potential hormone mimic/anti-hormone <sup>3</sup>			
setae/crochets	Х		mechanical/chemical abrasion of mucous membranes			
setae/setal fragments	x <sup>2</sup>	х	antigen/immunologic			
pathogen vector	<b>x</b> <sup>2</sup>	х	infection			
<sup>1</sup> Large numbers of caterpillars may need to be ingested.						

May be ingested along with grass or other forage passively

contaminated by dispersing caterpillars.

<sup>3</sup> Uninvestigated possibility.

the soft setae of the ETC are not designed to penetrate skin, and they are nonpoisonous. Yet, like the hairs of the processionary, they appear capable of causing an allergic skin response in individuals who handle large numbers of them over extended periods, but there have been no scientific studies to document these effects.

The last instar of the ETC accumulates calcium oxalate in its Malpighian tubules, structures that are the analogs of the kidneys of vertebrates. The ingestion of calcium oxalate can cause inflammation of the lining of the stomach and intestines, but it is not known if the small amount of the material found in the Malpighian tubules of the tent caterpillar has any significant effect on animals that ingest them. The caterpillar adds the oxalate to its cocoon while spinning, and it serves to stiffen it. If the cocoon is disturbed, the oxalate billows up in a yellow cloud and is thought to act as a deterrent to would-be predators. Anecdotal reports indicate that inhalation of the powder can irritate the respiratory tract, and contact with mucous membranes or skin may cause symptoms of redness, swelling, itching, and pain in susceptible individuals, but there have been no definitive studies to document any of these effects.

The ETC stands at the pinnacle of sociality among caterpillars. Caterpillars explore the branches of the host tree in a search for food. Successful foragers mark their pathways back to the tent with a recruitment pheromone that serves to guide other caterpillars to their food-find. Two pheromone components have been identified, and both are ketosteroids. There is the uninvestigated possibility that these compounds may have hormonal or anti-hormonal activity when consumed by predators. But because they occur in nanogram quantities, it would seem unlikely that they would have a significant impact on an animal as massive as a horse.

There is also the possibility that if a horse ingests a caterpillar, some attribute of the caterpillar may facilitate the invasion of potential pathogens that ordinarily reside harmlessly in the digestive tract of the horse. The setae or the sharply pointed crotchets of the prolegs could irritate the mucous membranes of the gastrointestinal tract of a vertebrate predator and create an infection court. A toxin produced by the caterpillar might act in much the same way.

Although no species of caterpillar is presently known to serve as a vector of a vertebrate pathogen, nor has any pathogen been thus far implicated in MRLS, a reasonable working hypothesis is that the ETC serves as a vector for a microbe that is a causal agent of MRLS. A good model species for such a pathogen is the bacterium Serratia marcescens, both because it has been isolated from the gut of field-collected caterpillars (5) and because it is capable of causing serious infection in horses (6,7). The bacterium has been little studied in tent caterpillars, but investigations of the tobacco budworm (8) indicate that it can be transferred from one generation of the caterpillar to the next on or in the egg of the moth. Moreover, the bacterium responds by multiplying rapidly when host caterpillars are stressed. The bacterium is typically benign in the gut of caterpillars, but if it gains access to the hemocoel, it causes rapid death (5). In species that can be killed by ingestion of the bacterium, oral doses of from 105 to 10<sup>6</sup> bacterial cells are typically required to produce significant mortality, indicating that common strains have low virulency. More virulent strains of S. marcescens, however, have been isolated. Farrar et al. (9) studied a strain that caused mortality in excess of 70% when fewer than 100 bacterial cells were fed to the corn earworm. Thus, studies of S. marcescens show that particularly virulent strains of viruses or bacteria may arise from time to time within populations of caterpillars and multiply rapidly when the insects are at peak population levels and stressed by starvation. Because of their enhanced virulence and abundance, these biological agents might pose a particular threat to cross-sensitive animals that contact the dispersing insects or their contaminated fecal pellets. Moreover, if the presence of virulent biological agents in populations of ETC is both a temporally and spatially transient phenomenon, the co-occurrence of ETC and pregnant mares need not invariably result in reproductive losses.

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# Age-Specific Patterns of Eastern Tent Caterpillar Dispersal: Implications for Reducing Mare Reproductive Loss Syndrome through Population Management

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THE EASTERN TENT CATERPILLAR (ETC), MALACOSOMA americanum (Fabricius) (Lepidoptera: Lasiocampidae), has been implicated in Mare Reproductive Loss Syndrome (MRLS) (1). This native defoliating insect overwinters as eggs in bands encircling small diameter twigs, and initial egg hatch coincides with bud break in early spring. Caterpillars feed gregariously and prefer wild cherry foliage (2), which grows abundantly in Kentucky along pastures and roadsides. Larvae construct conspicuous silken tents that increase in size as the caterpillars mature (2,3). At endemic population levels, caterpillars forage only as far as necessary to feed (4,5). But as populations increase to outbreak levels and preferred hosts are defoliated, host plant requirements become less stringent (6,7), and caterpillars are forced to forage greater distances to obtain food, thereby increasing the potential for exposure to grazing horses. ETC dispersal behavior has been studied extensively in the context of diurnal feeding patterns (4,5), trail pheromone production (8), and recruitment behavior (9), but nothing is known about how caterpillars disperse in pasture situations, nor how their dispersal behavior might be manipulated to affect population suppression.

Grazing restrictions, a limited treatment window, the potential for inclement spraying conditions, and environmental concerns make widespread insecticide applications for tent caterpillar control impractical on horse farms (10,11). Highly mobile, dispersing caterpillars and use of alternative trees by feeding caterpillars make efforts at focused control difficult. The ability to predict population behavior and develop more precise management strategies will be enhanced by a more complete understanding of age-specific caterpillar dispersal behavior and potential.

Caterpillar dispersal behavior was studied as it pertains to pasture situations in the Bluegrass region. The goal was to investigate the dispersal patterns of ETC from defoliated fence rows to potential alternative hosts and to potential overwintering sites. Specific objectives were to assess the dispersal patterns of foraging third and fourth instar ETC and compare them to dispersal patterns of wandering (late instar) caterpillars, with respect to dispersal direction, dispersal distance, response of caterpillars to visual stimuli, physiological capacity of caterpillars to disperse, and determining if the time of day influenced caterpillar dispersal behavior.

# Methods

Experiments were conducted in the spring of 2002 using field-collected ETC that were collected by clipping intact tents from wild cherry trees in and around Lexington, Kentucky. As they matured and local sources were depleted, additional caterpillars were obtained from south-central Wisconsin. Caterpillars were held in the laboratory in growth chambers (23°C, 15:8 L:D), fed fresh wild cherry foliage as needed, and were starved for 12 hours prior to use. All caterpillars were sorted and aged (12), and only those that appeared healthy were used in assays. The study site was a ~1.5 hectare, roughly rectangular asphalt parking lot on the University of Kentucky campus.

# Dispersal Distance

Caterpillars (n = 75) were released from the center point of the study site at 6 a.m. and 6 p.m. on three consecutive days (15 to 17 May 2002). There were three replicates of the 6 a.m. release, but because of inclement weather, there were only two completed replicates of the 6 p.m. release. Prior to their release, caterpillars were dyed with a fluorescent powder to facilitate tracking their movements. Caterpillar dispersal distance was monitored at 30, 60, 90, 120, and 360 minutes for those released at 6 a.m. and at 30, 60, 90, and 120 minutes for those released at 6 p.m. Evening releases could only be tracked for 2 hours because of darkness.

# **Dispersal Direction**

To assess caterpillar dispersal direction, arenas consisting of 20 m diameter circles were drawn in chalk on the asphalt. There were four blocks, each containing four circular 20 m arenas, separated by a minimum of 10 m, for a total of 16 arenas. Caterpillars (n = 25) were released from the center of each arena, and dispersal direction was monitored for 120 minutes. When caterpillars reached the edge of the circular arena, they were removed from the study. Measurements were made of the final bearing from the release point for caterpillars that did not leave their arenas.

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#### Response to Visual Stimuli

Caterpillar reliance on visual stimuli for orientation was assessed in two ways. First, caterpillar stemmata were blocked with acrylic artists' paint, effectively reducing all visual stimuli. The altered caterpillars (n = 25) were then released in the center of each circular arena (n = 16), and dispersal direction was monitored for 60 minutes. Secondly, response to visual cues was assessed by measuring the orientation of sighted caterpillars to vertically oriented objects, designed to simulate tree-trunk images. Each circular arena contained vertically oriented objects consisting of two adjacent 122 cm sections of 25 cm diameter PVC pipes, one of which was painted black and the other of which was painted white, placed at the edge of the arena in a randomly assigned direction. Caterpillars (n = 25) were then released in the center, and orientation and movement was monitored for 60 minutes. Again, when caterpillars reached the edge of the circular arena, they were removed from the study.

# Physiological Capacity of Dispersing Caterpillars

To assess the physiological ability of ETC to disperse, caterpillar stamina was tested on a treadmill. A small branch of host foliage, placed just out of reach of the experimental caterpillars, provided stimulus. Caterpillars were weighed prior to each trial, then placed individually on the treadmill (Model 1010-M3, Columbus Instruments, Columbus, OH) for 15, 30, and 120 minutes (n = 3), or for 240 minutes (n = 1). Preliminary experiments showed that caterpillars were easily able to travel at a rate of 2.7 m/ minute. Treadmill speed was set at that rate for all replicates except the single replicate lasting 240 minutes, during which the speed was reduced for some portions of the trial. Following each trial, the caterpillar was immediately frozen and stored at -80°C for future lipid analysis. Distance traveled was then calculated.

# Data Analysis

Analysis of variance was used to determine if distance traveled by released caterpillars differed between time increments, and linear regression was used to assess the relationship between distance traveled and time elapsed following release. After an analysis across all releases, a separate analysis for morning and evening releases was performed. Dispersal distance at each time increment (up to 120 minutes) for morning and evening releases was compared using a t-test. To assess the extent to which caterpillar dispersal direction was random, a chi-square analysis was performed separately for foraging and wandering caterpillars, for time of day (morning versus evening release), and for dispersal direction based on time elapsed (2 hours versus 6 hours post-release). To measure the randomness of orientation of caterpillars with blocked stemmata, a chisquare analysis was performed to assess the reliance of dispersing caterpillars on visual stimuli. Finally, a paired ttest was performed to assess differences in sighted caterpillar response to black and white vertically oriented objects.

# Results

# Dispersal Distance

Distance traveled by released caterpillars increased steadily and significantly (F = 23.7, P < 0.001), and was strongly correlated with the time elapsed in both the morning (F = 13.4, P = 0.003) and evening (F = 11.1, P = 0.001) (Figure 1) (F is a variance ratio test). Caterpillars released in the morning traveled at a fairly slow and steady rate of ~0.025 m/minute, equivalent to about 1.2 inches per minute, and covered an average distance of 8.4 m in 120 minutes. In the morning, the distance caterpillars traveled at 30 minutes differed from the distance traveled at 90 minutes (P = 0.02) and 120 minutes (P = 0.003), and the distance traveled at 60 minutes differed from that at 120 minutes (P = 0.03). However, there was no difference in distance traveled between the 30-minute and 60-minute time increments, between the 60- and 90-minute increments, and between the 90- and 120-minute increments. In contrast, the distance traveled by caterpillars released in the evening increased steadily but was similar across intervals. Caterpillars released in the evening traveled at a relatively rapid average rate of 0.17 m/minute (6.3 inches per minute) and covered an average distance of 39.3 m in 120 minutes, more than 4.5 times the distance of those released in the morning. The distance traveled by caterpillars released in the evening was greater than the distance traveled by caterpillars released in the morning at each time interval (Figure 1).

**Figure 1.** Distance travelled over time by ETC released from a central release point. Asterisks indicate significant differences between morning and evening releases at specified time intervals.



# Dispersal Direction

ETC dispersal direction was non-random and biased away from the south (Table 1a). This was particularly evident with foraging caterpillars, whose directional movement was strongly non-random and was greatest toward the northwest, west, and north. Dispersal direction of the older wandering caterpillars was random and differed significantly from directional patterns of the younger foraging caterpillars ( $\chi^2 = 47.3$ , P <0.0001).

The non-random dispersal pattern, biased away from the south, was more apparent in the morning (Table 1b), with caterpillar movement in the evening releases being only slightly non-random. The bias away from southerly movement persisted for up to 6 hours after the morning releases (Table 1c).

## Response to Visual Stimuli

Non-random dispersal persisted even when caterpillar stemmata were blocked, which effectively eliminated visual stimuli. However, the pattern differed significantly from sighted caterpillars ( $\chi^2 = 131.9$ , P <0.0001). In altered caterpillars, the bias in caterpillar orientation shifted, with the lowest response in the northwesterly direction (Table 1d).

Sighted caterpillars were responsive to vertically oriented objects, and when given a choice, showed a strong preference for black objects over white (Table 2).

# Physiological Capacity of Dispersing Caterpillars

Caterpillars readily moved on the treadmill for 15-, 30-, and 120-minute intervals, traveling distances of up to 324 m (Table 3). The single caterpillar that lasted 240 minutes required some light prodding and eventually stopped from exhaustion after traveling a distance of approximately 624 m. feeding activity (5). Caterpillar dispersal rate is temperature dependent and is a function of body length and body temperature (13). The air temperature at 6 a.m. was 12.3°C, with an average of 15.2°C over the 120-minute morning assay. At 6 p.m., the air temperature was 17.1°C, averaging 17.9°C over the corresponding 120 minutes. Although surface temperature measurements were not consistently taken over the course of the study, 6 a.m. surface temperature averaged 19.5°C over two measurements, and 6 p.m. surface temperatures were 27°C, also averaged over two measurements. Clearly the elevated evening temperatures could influence caterpillar dispersal rate.

Dependence of ETC on visual stimuli for orientation is evidenced by the shift in caterpillar movement when visual cues are obstructed. Polarized light, detected through caterpillar stemmata, may be used for orientation. Wellington (14) demonstrated that by rotating the plane of polarized light with polarizing filters a specified amount, tent caterpillar movement can be shifted by a similar amount. The shift in caterpillar movement when stemmata are obstructed supports the idea that polarized light may serve as a cue for caterpillar orientation. Caterpillar stemmata also detect light and allegedly are capable of crude image formation (15). Larvae swing their heads back and forth, scanning their surroundings, and can detect shapes and object orientation (16,17). Studies of other treefeeding lepidopterans have demonstrated caterpillar orientation toward objects (18) and a limited ability to discern an object's size and relative distance (19). The results corroborate these findings and demonstrate that the ETC is able to detect the contrast between black and white and may be capable of limited color vision.

## Discussion

During the morning hours, foraging ETC do not disperse randomly but show a bias away from southerly movement that is maintained for up to 6 hours following release. In contrast, foraging caterpillars released in the evening were more random in their dispersal direction, moving in all directions somewhat evenly. The magnitude of the differences in distance traveled between the morning and evening releases is striking since both releases approximate periods of tent caterpillar

<b>Table 1.</b> Dispersal direction (% response) of ETC released under various conditions from the	2
center of 20 m diameter circular arenas (n = 16).	

		Compass Direction								
		Ν	NE	E	SE	S	SW	W	NW	χ²/ <b>Ρ</b>
a.	Caterpillar age									
	all caterpillars	20	16	13	8	8	8	14	14	42.2/ <0.001
	foraging	18	11	11	3	4	6	19	28	302.4/ <0.0001
	wandering	16	15	14	10	10	9	11	16	11.6/ 0.11
b.	Release time									
	morning	20	19	14	7	6	7	12	16	43.7/ <0.001
	evening	19	11	12	10	12	9	17	11	12.2/ 0.09
c.	Time elapsed									
	2 h post-release	19	13	13	9	9	9	15	14	19.7/ 0.006
	6 h post-release	21	22	13	6	6	6	11	13	32.9/ <0.001
d.	Visual status									
	sighted 20		16	13	8	8	8	14	14	42.2/ <0.001
	altered	12	14	16	13	14	13	11	7	26.8/ 0.004
а	Caterpillar age: "fora	aging" aı	re third	and fou	ırth inst	ar cate	rpillars, a	and "w	anderin	g" are fifth and sixth

instar caterpillars, independent of release time and time elapsed.

<sup>b</sup> Release time: 6 a.m. and 6 p.m., independent of caterpillar age and time elapsed.

c Time elapsed: 2-hour and 6-hour post-release, for morning releases only, independent of caterpillar age.

d Visual status: "sighted" are caterpillars with complete vision, and "altered" are caterpillars with stemmata obstructed.

Table 2. Response of foraging ETC to
black and white vertically oriented
objects.

Color	Mean (s.e.)
black	1.7 (0.2)
white	0.5 (0.1)
paired t-test / P	5.3/ <0.0001

Future work on ETC dispersal as it pertains to MRLS will concentrate on developing a model to predict caterpillar location (distance and direction) based on time of day (morning versus evening) and time elapsed since caterpillar movement began. Preliminary work using a logit model (20) demonstrated that caterpillar distance and direction can be predicted based on time of day and time elapsed, but the existing model lacks precision.

Knowledge of caterpillar dispersal patterns will increase our ability to manage ETC populations and predict risks to specific pastures. In addition, this knowledge will help in the development of management protocol, such as isolation distances of mares from host trees, that will allow horse farm managers to more effectively reduce mare exposure to tent caterpillars, thereby reducing the risk of MRLS.

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Table 3. Distance traveled by foraging ETC in treadmill studie	s.
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Rate (m/minute)	Time Elapsed (minutes)	Distance Traveled (m)	n
2.7	15	40.5	3
2.7	30	81	3
2.7	120	324	3
2.5-2.7	240 <sup>a</sup>	624	1

<sup>a</sup> to exhaustion.

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# Timing of Emergence of Eastern Tent Caterpillars and Management with Reduced Risk Insecticides and Treatment Strategies

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FARM SURVEYS (1, THIS PROCEEDINGS) AND EXPERIMENTAL induction of Mare Reproductive Loss Syndrome (MRLS) by directed exposure of susceptible mares to eastern tent caterpillar (ETC), Malacosoma americanum, in field plots (2, this proceedings) or by gastric administration (3, this proceedings) point to ETC being involved with the disease. This mandates that farms minimize exposure of pregnant mares to ETC at least until the mechanism by which the caterpillars are involved can be determined. Spraying tall trees bordering pastures with traditional insecticides is potentially hazardous to farm workers, horses, and the environment. Moreover, relatively few insecticides are labeled for pasture usage, and many products that are effective against ETC have grazing restrictions on their label. Issues of spray drift and liability thus complicate control options.

Reduced-risk insecticides and treatment strategies for managing ETC on horse farms were evaluated. The work emphasized target-selective compounds with low vertebrate toxicity, or micro-injection of trees to avoid spray drift. ETC egg hatch was monitored to help guide the timing of control actions. The 2002 results of these ongoing studies are summarized.

# **Materials and Methods**

## Timing of Emergence in Trees

Emergence of ETC from egg masses and subsequent colony development were monitored at three field sites, the University of Kentucky's Coldstream and South Research Farms and Gainesway Farm, near Lexington, Kentucky, in the spring of 2002. Sites were rows of mature wild cherry trees, *Prunus* spp., bordering pastures or fences. About 200 total twigs bearing egg masses were tagged with flagging tape from 11 to 19 February. Egg masses were checked every 1 to 2 days until mid-April, when all larvae had emerged. Larval behavior (e.g., aggregation on egg masses, movement to twigs, size of nests) and instars predominating were noted. Observations continued until larvae began wandering before pupation.

Twenty additional cherry twigs with single ETC egg masses were field-collected and kept outdoors in florists' water picks to maintain shoot turgor. These egg masses were monitored daily; upon eclosion, larvae were counted and removed. The pattern and duration of emergence from individual egg masses were determined, as well as the number of ETC per mass.

# Horticultural Oil or Insecticidal Soap against Newly Hatched Larvae

Insecticidal soaps and oils seemingly were good candidates for ETC management because they are essentially nontoxic to vertebrates. Egg masses with newly emerged and soon-to-emerge larvae were collected from field sites. Twigs bearing egg masses with aggregations of newly enclosed larvae were sprayed to runoff with either a 3% solution of Superior miscible oil (Universal Cooperative, Minneapolis, MN), insecticidal soap (M-pede<sup>®</sup>, Mycogen, San Diego, CA) at labeled rate (31.3 ml/liter [4 fl oz/gal.]), or distilled water. Another set of controls was not sprayed. After treatment the larvae (about 25 per egg mass) were brushed off the twigs onto moist filter paper in separate petri dishes for each mass. Mortality was evaluated after 1, 4, 8, 24, and 48 hours. Data were adjusted for control mortality by Abbot's formula (5).

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#### Foliar Sprays for Controlling Early or Late Instars

Four insecticides with low mammalian toxicity were tested against young (first and second instar) or midsized (third and fourth instar) ETC feeding on fresh cherry foliage. These products were: Bacillus thuringiensis [Bt] (Dipel Pro®, Valent, Richardson, TX), a microbial insecticide active against many caterpillar species; Spinosad (SpinTor<sup>®</sup> 2SC; Dow Agrosciences, Indianapolis, IN), derived from fermentation of a naturally occurring soil bacterium (4); tebufenozide (Confirm® T/O, Dow AgroSciences, Indianapolis, IN), a molt-accelerating compound (ecdysone agonist); and bifenthrin (Talstar®, FMC, Philadelphia, PA), a pyrethroid. Each product was applied at label rate: Dipel at 2.64 ml/liter [34 fl oz/100 gal.]; SpinTor at 0.66 ml/liter [8 fl oz/100 gal.], Confirm at 1.32 ml/liter [16 fl oz/100 gal.], and Talstar at 0.84 ml/ liter [10.8 fl oz/100 gal.]. A spreader/sticker (Breakthru®) was added to the tank mix at 0.31 ml/liter [4 fl oz/100 gal.] for all spray tests.

Tests with early instars were done twice, with woody shoots bearing either green-tip buds or expanded leaves, to simulate conditions encountered at emergence. Tests with larger larvae used fully expanded leaves. Larvae and shoots were field-collected from wild cherry trees. Shoots were trimmed to 25 cm length, placed temporarily in water-filled flasks, sprayed to runoff, with residues allowed to dry outdoors. Separate controls were sprayed with the spreader/sticker only or were not treated. Individual shoots then were inserted through a white 10-cm filter paper and the slotted lid of a water-filled plastic cup. A vented, acrylic cylinder was placed over each shoot to form a cage. The paper floor facilitated counting of moribund or dead larvae, as well as collection of frass. Treatments were replicated five times in each trial. Treated shoots were challenged with 20 ETC for tests with early instars or with 10 ETC for tests with third and fourth instars. Caged shoots were held in a growth chamber set to simulate concurrent field conditions: 9:15 (L:D) photoperiod, and 25°C or 15°C, respectively, during those cycles. Mortality of early instars was evaluated after 1, 8, and 24 hours, and daily thereafter for 7 days. Mortality of midsized ETC was determined daily for 7 days. Accumulated frass was collected, dried, and weighed.

#### Toxicity of Weathered Residues on Foliage

Persistence of the aforementioned insecticides was tested as follows. Pre-tagged cherry shoots were sprayed on 14 April, with residues allowed to weather in the field. Separate sets of shoots were harvested at 1, 3, or 7 days after treatment and challenged with third and fourth instars as above. Larval mortality was assessed daily for 4 days after each respective harvest.

## Test for Behavioral Avoidance of Residues

It was important to determine if residues of the aforementioned insecticides might repel ETC because such response might cause larvae to avoid feeding on treated leaves or even to prematurely abandon treated trees and disperse into pastures. Shoots with fully expanded cherry leaves were treated on 30 April with residues allowed to dry. Treated leaves were paired with nontreated ones in 10-cm petri dishes on moist filter paper. There were 30 replicates for each insecticide. One fourth-instar ETC was added to each dish and left for 3 hours to take its first sustained meal (typically 10 to 20% of one leaf was consumed). Leaves then were electronically scanned, and missing leaf areas were compared.

# Micro-Injection of Cherry Trees with Dicrotophos (Bidrin)

Mature cherry trees growing along a fence at Glenwood Farm, Versailles, Kentucky, were used in this study. The trees were heavily infested with many small tents containing mostly second-instar ETC. The trial was done on 10 trees in a continuous row. On 16 April, alternating trees were injected with dicrotophos (Bidrin, as Inject-A-Cide® "B") using the Mauget Micro-Injection System (Mauget, Los Angeles, CA). Dosage was determined by dividing tree circumference (in cm) at 1.25 m above ground by 15 (e.g., a 22.5-cm diameter tree received five capsules). Injections were done in the root flare, about 15 cm above ground, and evenly spaced around the tree. Control trees were not injected. Efficacy was determined after one week (23 April) by harvesting 10 nests per tree (50 nests per treatment) with a pole pruner or by climbing and sealing them in individual bags. Most of the sampled nests were 4 to 8 m above ground. Accurate whole-tree nest counts were impossible because the trees were flowering, and small tents could not be seen against the backdrop of white flowers. Each control tree was injected immediately following the 23 April sampling. Those trees were sampled again, in similar manner, on 30 April.

Bagged nests were taken to the laboratory where they were dissected and the caterpillars examined. Nests initially were scored: 1 =all larvae dead, 2 =a few ( $\leq 5$  live larvae), 3 = 5 to 10 live larvae, 4 = 25 to 50 live larvae, 5 =robust, full nest. All 50 nests from the treated trees were then dissected, their larvae individually examined and counted. Nests from control trees all were robust, and many contained hundreds of live larvae. For controls, a representative sample (10 nests; two from each tree) was dissected as above.

# *Test for Residual Control of Wandering Larvae in Pastures*

This trial was conducted in a mixed bluegrass pasture at the University of Kentucky's Spindletop Research Farm, near Lexington. Individual plots (6.4 x 6.4 m) were arranged in a randomized complete block with four replicates. Treatments were Malathion 25% EC at 1.4 kg AI/ha [1.25 lb AI/A], Sevin 4F at 1.12 kg AI/ha [1 lb AI/A], or untreated controls. Plots were sprayed on 7 May. When residues had dried, two rings of polyvinylchloride (PVC) pipe (15.2 cm high, 20.3 cm diameter) were seated about 1 m apart near the center of each plot. Ten fifth-instar ETC, collected from leaves of wild cherry trees, were placed inside each ring, the top 2.5 cm of which was greased with petroleum jelly to discourage escape. Plots were examined on May 8 and 10, and numbers of live and dead larvae were determined.

# Preliminary Test for Residual Toxicity of Bifenthrin to ETC on Tree Trunks

The final test, done 14 May, examined whether dry residues of bifenthrin (Talstar) or permethrin (Astro<sup>®</sup>, FMC, Philadelphia, PA) on cherry bark might intercept and kill ETC crawling down tree trunks to disperse or pupate. Fifteen freshly cut sections of cherry trunk (2 m long, 15 to 20 cm diameter) were obtained from a tree care firm. These were supported vertically, out of doors, and treated with either Talstar (7.9 ml/liter [1 fl oz/gal.], Astro (13.2 ml/liter [1.7 fl oz/gal.), or not treated. When residues had dried, 10 late-instar ETC were placed on the top of each bolt, and their behavior was observed. Two separate groups of larvae were tested with each bolt.

# Results and Discussion

# Timing of Emergence in Trees

First observed emergence from egg masses was 15 March 2002, coincident with about 50% bloom of *For-sythia*. Once an egg mass became active, neonates continued to emerge for two to three weeks (mean =  $17.9 \pm 1.0$  day, range 12 to 26 days). Area-wide emergence was slow during the first 1.5 weeks but accelerated as mean daily temperatures climbed. Emergence peaked from 29 March to 2 April and was nearly finished by 15 April. Only 44% of the egg masses had become active by 28 March, whereas 88% had done so by 3 April. Larvae did not begin to emerge from the last egg mass until 18 April. Small tents were visible in cherry trees by 1 April, and by 10 April many larvae were abandoning smaller nests on the tree periphery to join together into larger central nests in main tree crotches.

# Horticultural Oil or Insecticidal Soap against Newly Hatched Larvae

Point-blank sprays with horticultural oil or insecticidal soap gave poor control of neonate ETC. Adjusted percentage mortality from those treatments averaged  $27.2 \pm 6.7$ versus  $38.1 \pm 3.1$ , respectively, after 24 hours, and  $37.1 \pm$ 4.7 versus  $49.1 \pm 5.2$ , respectively, after 48 hours. Given the prolonged emergence period of ETC and the fact that oils and soaps kill by contact and have no residual activity, these products would have to be sprayed repeatedly, every few days, to get even partial control. Efficacy doubtless would be even less once ETC had molted to second instars. Oils or soaps would be further compromised by the difficulty of getting adequate coverage in tree canopies.

# Foliar Sprays for Controlling Early or Late Instars

Dry residues of Talstar or SpinTor gave excellent control of first instars (100 and >95%, respectively, within 24 hours) on both green-tip buds and young leaves. Dipel also was effective but slower. Mortality of neonates gnawing on Dipel-treated green-tip buds was 31, 52, and 61% after 24, 48, and 72 hours, respectively. Corresponding levels of control on Dipel-treated expanding leaves were 43, 89, and 92%. Confirm, the molt-accelerating compound, worked more slowly than Dipel, providing only 45% control after 3 days, and about 75% control after 5 days. Talstar works both by contact and ingestion, whereas Spintor, Dipel, and Confirm must be consumed.

Against third and fourth instars, Talstar and Spintor gave 78 and 76% control, respectively, after 1 day; 96 and 100%, respectively, after 2 days; and both gave 100% control after 3 days. Confirm gave 66% control after 2 days, reaching 100% control after 4 days. Dipel was slower against third and fourth instars than against younger larvae, providing 6, 66, and 84% mortality after 1, 4, and 5 days.

## Toxicity of Weathered Residues on Foliage

Feeding of midsized ETC on cherry leaves with 3-dayold weathered insecticide residues resulted in 100, 73, 39, and 20% mortality for Talstar, Spintor, Dipel, and Confirm, respectively, after 48 hours. Similarly, feeding on 7-dayold residues of those products gave 100, 59, 23, and 16% mortality, respectively, after 48 hours. Talstar clearly worked fastest and had the longest residual effectiveness of the insecticides we tested. For example, 3-day-old residues of Talstar gave 80% control after 1 day, and 100% within 2 days. Mortality for larvae fed similar-aged residues of Dipel (Bacillus thuringiensis) was 6, 39, 48, and 81% after 1, 2, 3, or 4 days, respectively. Larvae fed 7-dayold Talstar residues suffered 47 and 100% mortality within 1 or 2 days, respectively, whereas 7-day-old Dipel residues gave only 6, 23, 35, and 52% mortality after 1, 2, 3, or 4 days, respectively.

## Test for Behavioral Avoidance of Residues

ETC showed no behavioral avoidance of residues of any of the insecticides. Numbers of pairings (counting only those in which the larva fed) wherein the treated leaf had more damage than the control were 5 of 9 for Talstar, 13 of 25 for Dipel, 14 of 24 for Spintor, and 11 of 20 for Confirm. The low number of total pairings for Talstar reflects the relatively high mortality in that treatment.

# Micro-Injection of Cherry Trees with Dicrotophos (Bidrin)

Micro-injection with bidrin was very effective when the treatment was done early (16 April), when most (about 90%) of the ETC were second instars and the tents were small. All 50 nests harvested from control trees were viable and robust. Those nests contained a mean ( $\pm$  SE) of 304  $\pm$  91 live larvae each (range 47 to 978). Of the 50 sampled nests from treated trees, 33 contained all dead larvae, and the rest contained just a few survivors. Collectively those 50 nests contained only 136 live larvae. The 16 April treatment therefore gave about 99% control.

The ETC population was developmentally more advanced when the original control trees were injected on 23 April. At that time, the mean proportions of second, third, and fourth instars present were 17, 63, and 20%, respectively. The 50 nests sampled from those trees on 27 April contained means of 190  $\pm$  29 dead or moribund ETC versus 266  $\pm$  37 active larvae, representing about 42% control. Larvae were mostly fourth and fifth instars by that time.

# *Test for Residual Control of Wandering Larvae in Pastures*

Treating pasture grass with Sevin or Malathion gave almost no residual control. Numbers of live larvae recovered after confinement on freshly treated grass for 24 hours were  $15.5 \pm 0.9$ ,  $13.7 \pm 1.7$ , and  $12.8 \pm 1.7$  for control, Sevin, and Malathion, respectively. Corrected mortality, based on the proportion of dead versus live larvae, was <5% for each treatment. Totals of 50, 47, and 44 live larvae were recovered from those plots, respectively, after 72 hours.

# Preliminary Test for Residual Toxicity of Bifenthrin to ETC on Tree Trunks

This method appeared ineffective. ETC crawling from the tops of the cherry trunk sections (where they had been placed) invariably dropped to the ground as, or before, they encountered the insecticide-treated bark and appeared unaffected. The assay may not have realistically simulated behavior of larvae that are behaviorally predisposed to crawl down tree trunks. However, the outcome is consistent with observations of ETC dropping from, as well as crawling down, trees in the field.

# Conclusion

The prolonged emergence period of ETC complicates its management with short-residual insecticides. Soaps and oils, which have no residual, must be sprayed directly on target insects and therefore are ill suited to this problem. They do not penetrate the silken nets, and getting adequate coverage in tree canopies is problematic. *Bacillus thuringiensis* (Dipel) works well against small ETC but is slower and less effective against late instars. Because it has relatively short residual activity, three weekly applications, starting a few days after first egg hatch, likely would be needed for a high degree of control.

Talstar (bifenthrin) was the most effective, fastest, and longest-lasting foliar insecticide that we tested. It controlled both small and large larvae, so by waiting until most of the ETC population had emerged (two to three weeks after first egg hatch), farm managers likely could obtain excellent control with one application. SpinTor (spinosad) was nearly as effective, but label restrictions (see below) may limit its versatility for horse farms.

Both the Environmental Protection Agency's (EPA) 1992 Worker Protection Standard and label grazing restrictions limit the range of insecticides that can be applied to trees on horse farms. Dipel can be used on such sites, with no grazing restrictions. Of the several bifenthrin formulations, Talstar Nursery Flowable is labeled for agricultural use sites, including trees on horse farms (G. Meinke, personal communication). It carries no grazing restrictions (but it is not labeled for direct application to pastures). Grazing restrictions on spinosad labels (Conserve<sup>®</sup>, SpinTor<sup>®</sup>) limit its application to sites where drift onto pastures will not occur. Farm managers and applicators should read labels and consult regulatory officials to ensure compliance with label restrictions.

Micro-injection of trees with bidrin was highly effective against young ETC but less so against larger larvae. Bidrin is quite toxic to mammals, but micro-injection by a certified arborist allows it to be delivered systemically, with no drift and low hazard to livestock or farm workers. Studies planned for 2003 will evaluate less inherently toxic systemic insecticides (e.g., abamectin) and delineate the temporal window during which such applications are effective.

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# Use of Recommended Insecticides Tebufenozide and Spinosad in the Vicinity of Broodmares

#### K. McDowell

IN THE EPIDEMIOLOGICAL SURVEY OF 133 KENTUCKY horse farms following the 2001 Mare Reproductive Loss Syndrome (MRLS), presence of black cherry trees and eastern tent caterpillars (ETC) were significant risk factors for mares that lost their pregnancies (1). Thus, by late summer of 2001, the University of Kentucky College of Agriculture recommended control of ETC for reducing or preventing MRLS in the future (2,3). Two of the insecticides, tebufenozide (Confirm®, Rohm & Haas, Philadelphia, PA) and spinosad (Conserve<sup>®</sup>, DowElanco, Indianapolis, IN), were relatively new products and as such may not be as familiar to horse farm owners and managers as some of the more traditional insecticides. Therefore, this project was initiated to address questions from local horsemen about the use of these newer insecticides in the vicinity of horses.

Tebufenozide and spinosad are biologically active insecticides and are recommended for control of ETC (3). The insecticidal activity of both of these products is expressed primarily through ingestion by caterpillar larvae. They are designed to be sprayed onto tree leaves on which the caterpillars feed. Thus, the question was raised regarding possible wind drift of the insecticides and hence accidental ingestion of small amounts of the products by grazing horses.

No reports were found in the literature where these insecticides had been used specifically in the vicinity of horses, and at the time this project was conducted (February of 2002), neither product was approved for use on pasture or with grazing animals. However, for the sole purpose of this research project, the insecticides were applied directly onto the hay fed to pregnant mares.

# **Materials and Methods**

Sixteen pregnant mares, approximately 250 days gestation, were divided into two groups of eight mares each. Both groups were maintained in adjacent pastures and supplemented with hay fed on the ground. Tebufenozide and spinosad were mixed together with water in a 2-gallon sprayer, at rates suggested by the manufacturers. The insecticides were sprayed directly onto the hay fed to one group of eight mares. Water was sprayed onto the hay fed to the second group of eight mares. Hay was sprayed to the point that the hay was slightly dripping. It was treated on Monday, Wednesday, and Friday for one week (three treatments total).

All mares were palpated, their pregnancies examined by ultrasonography, and blood samples were taken before treatment started and 8 and 18 days after treatment started. Serum samples were analyzed for total estrogens, progesterone, and thyroid hormone (T4) (Bluegrass Embryo Transplants, Lexington, KY) and clinical chemistry (the University of Kentucky Livestock Disease Diagnosis Center [UKLDDC]). Data were analyzed by the Mixed Procedure of the Analysis of Variance (6) for main effects of treatment and day and the treatment by day interaction.

#### Results

The mares readily ate hay that was sprayed with either insecticides or water. No adverse effects were observed for the mares or for their pregnancies. All mares had normal healthy foals. All blood chemistry values were within

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acceptable ranges. Average glucose concentrations increased over time, but there were no differences between treatment groups (Table 1). Serum hormone concentrations are shown in Figure 1. Estrogen concentrations were higher on January 31 than on February 11 or February 21, but there were no differences between the treatment groups (Table 1).

## Discussion

There is increasing evidence that MRLS is caused, in whole or in part, by ETC. Several methods are recommended for control of these insects, including spraying insecticides on trees on which the caterpillars are feeding (2,3). Many people are interested in safer, more environmentally friendly insecticides than those frequently used in the past. Two of the newer insecticides recommended by the University of Kentucky College of Agriculture for the control of ETC are tebufenozide and spinosad (2,3). Both are targeted toward specific classes of insects, not mammals, and their reported toxicities in mammals are extremely low (4,5).

Tebufenozide induces caterpillars to molt. Insects periodically shed their outer cuticle layers in a process called molt. Molting is initiated by an increase in levels of the steroid hormone 20-hydroxyecdysone (20E). 20E binds to its receptor, ecdysteroid receptor protein, and activates genes responsible for the molting process. Molting is terminated by a decline in 20E to basal levels. Tebufenozide is a metabolically stable ecdysteroid receptor agonist, and thus it induces the molting process. The caterpillars are unable to terminate this induced molt and die of dehydration or starvation. All molting insects utilize the 20E/receptor complex, but molt inhibitors such as tebufenozide do not function similarly with all insects. This selectivity may be due, at least in part, to different binding affinities of the compounds for the different insect systems. In addition, 20E and its receptor are not found in mammals; thus, 20E agonists cannot serve as specific ligand agonists in mammalian systems (4,7,8). 20E agonists have, however, also been found to affect membrane structure and function, presumably via significant lipophilic activity (9). Indeed, they have been reported to produce excitotoxicity by blocking K+ channels (10). Thus, there may be potential for neurotoxicity, independent of 20E/receptor activity.

Spinosad is a mixture of two naturally occurring metabolites of *Saccharopolyspora spinosa*, spinosyns A and D (11). Spinosad targets the insect nervous system, affecting gamma-amino butyric acid (GABA) receptor function, and causes involuntary muscle tremors, paralysis, and death. Differences in insect and mammalian nervous systems account for the reported safety of this insecticide for mammals (11,12).

Table 1. Mean, standard deviation of the mean (Stdev	) and
standard error of the mean (SEM) for each day tested.	

	(	Glucose		Estrogens			
	Mean (mg/dl)	Stdev	SEM	Mean (pg/ml)	Stdev	SEM	
Jan 31	42	18	5	2464	687	172	
Feb 11	66	20	5	1848	601	150	
Feb 21	77	14	4	2116	550	138	
Significance values for pairwise differences between dates.							
Glucose Estrogens							
Jan 31 vs. Feb 11 p				0.0305	p < 0.	.0002	
Jan 31 vs. Feb 21		p < 0.0001		p < 0.0103			
Feb 11 v	s. Feb 21		p < 0.0550		p < 0.0542		

When these insecticides were sprayed onto hay that was fed to pregnant mares, no adverse effects were seen on the mares themselves, their *in utero* pregnancies, or on the foals that were born to these mares. All mares delivered live, healthy foals. Several of the foals have been weaned as of this writing and continue to be in good health.

Figure 1. Hormone concentrations in mares when hay was sprayed with insecticides (open bars) or water (shaded bars).



Both of these insecticides are designed to be sprayed onto the tree foliage where caterpillar larvae are feeding. They are ineffective if sprayed onto the caterpillar itself. They were only used on hay in this experiment as a direct test for effects on the horses, not as a means of caterpillar control. Package instructions should be followed when using any insecticide, and appropriate care should be taken to minimize human and animal exposure.

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# **Field Experiences with Caterpillar Control**

S. E. Johnson

RECENT STUDIES HAVE ESTABLISHED THE RELATIONSHIP BEtween Mare Reproductive Loss Syndrome (MRLS) and the presence of the eastern tent caterpillar (ETC). Although the exact mechanism of the loss of foals and pregnancies attributed to MRLS still remains unknown, from a management standpoint, we now have sufficient information to begin to implement control measures for MRLS by controlling ETC concentrations on our farms.

# Management Practices Initiated Prior to the Establishment of ETC as the Link to Mare Reproductive Loss Syndrome

There were several hypotheses as to the cause(s) of MRLS in 2001, and based on some of these theories, a great majority of the farms in Central Kentucky established contingency plans in 2002 in an effort to alleviate the high losses of early-term pregnancies and late-term foals that so many farms experienced in 2001. These contingency plans were based on what information was available to the industry after studying many of the factors of MRLS that were summarized in several surveys and field reports from 2001. Some of the areas into which researchers were looking were the ETC, mycotoxins, weather patterns, and how horses kept on pasture may be exposed to various viruses, bacteria, toxins, minerals, and other pathogens that could be the cause of MRLS.

Prior to the work that indicated the presence of ETC was responsible for MRLS, management practices were implemented on many horse farms in Central Kentucky early in the spring of 2002 in an effort to reduce losses to MRLS. These practices included:

- limiting the amount of time broodmares are exposed to pasture
- muzzling broodmares
- keeping broodmares in drylots
- spraying wild cherry trees with many different pesticides
- applying "sticky collars" around the trunks of trees
- attempting to pick out ETC egg masses from wild cherry tree branches
- "vaccinating" wild cherry trees
- burning ETC webs that were found in trees
- keeping pastures mowed as short as possible
- chain harrowing pastures
- · cutting down and removing wild cherry trees
- moving broodmares out of Kentucky
- moving broodmares to different locations on the farm
- delaying breeding mares until after the middle of March

- adding binders to feed rations
- making different mineral supplements available to broodmares.

# ETC Established as the Link to Mare Reproductive Loss Syndrome

As soon as it became apparent that ETC was the culprit, it became clear that many of the above practices had helped reduce the incidence of MRLS in 2002. Now breeders and managers could begin to have confidence in their efforts to limit exposure to ETC and not have to continue to employ such varied "shotgun" management techniques to control MRLS on their farms.

Currently we see farms taking strong measures to reduce exposure to ETC by removing the primary habitat of ETC, which is the wild cherry tree. These trees are being cut down, girdled, and killed with herbicides in an effort to control ETC.

# **ETC Control Measures**

Additional study and work is needed to find other means of control for ETC. The chemical controls, while somewhat successful, still have limitations because of the vast expanse of wild cherry trees throughout the region, the difficulty in spraying many of the larger wild cherry trees, the exponential concentrations of ETC during some years, the migratory nature of the caterpillar, and environmental and toxic effects of some pesticides. Even though many farms implement control measures within their borders, if their neighboring farms do not have ETC control practices, then many caterpillars migrate across property lines and cause problems to the farms that are working hard to control ETC on their property.

Several farms reported a great deal of success "vaccinating" their wild cherry trees with systemic pesticides. This procedure involves injecting a measured amount of a pesticide into the trunk of a wild cherry tree, and when this pesticide is transpired throughout the tree, the early larvae feed on the leaves of the treated trees. When the young larvae emerge from their egg mass and feed on the treated leaves, they soon die.

An obvious area that will require further research and study is with insecticides. Although many farms had success with spraying insecticides on ETC, it was reported that the most success was when the young larvae were still in

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the trees and had just begun to feed on leaves that were treated with pyrethrins and/or carbaryl products. Although insecticides in the organophosphorus family may be highly effective, they have been shown to be toxic to mammals.

# Conclusion

Now that we are aware of the connection between ETC and MRLS, and as helpful as it will be for researchers to

learn the mechanism by which ETC causes MRLS, it is of paramount importance that future work be directed toward ways to control ETC. Principally, efforts should be made by the entomologists and the chemists to find safe and effective means with which to control ETC. By having stronger and more predictable techniques to be better able to control ETC, managers and horsemen will be better able to control the incidences of MRLS on their farms in the future.

# Potential Uses for the Eastern Tent Caterpillar Sex Pheromone

## K. F. Haynes

SEX PHEROMONES MEDIATE MATING BEHAVIOR FOR MOST moths. Typically the female moth releases a pheromone blend that stimulates the male moth to fly upwind. The eastern tent caterpillar (ETC) moth, Malacosoma americanum, fits this pattern. Females release a blend of (E,Z)-5,7-dodecadienal and (E,Z)-5,7-dodecadienol. Males are attracted to a blend of these compounds, but more work needs to be done to define the most effective pheromone blend. Pheromones have been used to help control insect pests in a number of different ways, and some of these could be used against ETC moths. Pheromone traps could be used to monitor flights of male moths. This could be useful in mapping the geographical distribution of ETC or following the long-term population cycles that characterize this species. Pheromone traps could also be used to define the end of a seasonal flight period, which could be useful if a decision is made to remove host trees after egg laying has occurred. With other species, pheromones have been used for more direct manipulations of insect populations. The

trap-out tactic involves removal of enough males to limit reproduction. This approach is very labor intensive. Mating disruption involves wide dispersion of the sex pheromone resulting in inability of males to find females. Sometimes insecticides are combined with the pheromone to "attract and kill" males. Another possibility is to use pheromones as an aid in auto-dissemination of an ETC-specific virus. Males would be attracted to a pheromone source that contains a formulation of the virus. Males would subsequently contaminate females with the virus, who would contaminate the eggs. The success of tactics that use sex pheromones depends on the proportion of males affected, the area covered with the pheromone treatment, and the distance of migration of mated females. These methods have the potential to interrupt the annual life cycle of this insect, but action is required during the very short adult lifespan.

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# Summary

#### T. D. Fitzgerald

WE HAVE QUITE A DIVERSE GROUP HERE IN TERMS OF THEIR interests, and how does one summarize these different ideas? I think two things come to mind. As Dr. Webb said earlier, the kind of research that Dr. Rieske-Kinney is doing would never have been funded several years ago. If you went to a granting agency and said, "I want to find out how far a caterpillar walks," I don't believe you would have gotten much money. This becomes extremely relevant, as this kind of basic information is very important. The other concerns control. It seems to me that there shouldn't be an easier insect to control than the ETC. What we need is a laser gun to target the eggs and blast them out of the trees. Another possibility is a small capsule that you shoot into the tent. It would open, and a volatile chemical would come out and saturate. Those caterpillars will die because they come back to the tent, they love that tent, and they won't leave it. Why cut down the tree? Well, it's like throwing the baby out with the dirty bathwater. It's overkill. Appropriate technology may be a possibility here, and there's a good opportunity to develop it.

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