Case-control study of late-term abortions associated with mare reproductive loss syndrome in central Kentucky

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Objective—To identify factors associated with abortions of mares during late gestation attributed to mare reproductive loss syndrome (MRLS).

Design—Case-control study.

Animals—282 broodmares from 62 farms in central Kentucky, including 137 mares that had late-term abortions (LTAs) associated with MRLS, 98 mares from the same farms that did not abort, and 48 mares that aborted from causes other than MRLS.

Procedure—Farm managers were interviewed to obtain data on a wide range of management practices and environmental exposures for the mares. Data for case and control horses were compared to identify risk factors for a mare having a MRLS-associated LTA (MRLS-LTA).

Results—Several factors increased the risk of mares having MRLS-LTAs, including increased amount of time at pasture, less time in a stall, feeding concentrate on the ground, higher proportion of diet derived from grazing pasture, being fed in pasture exclusively during the 4-week period prior to abortion, access to pasture after midnight during the 4-week period prior to abortion, and drinking from a water trough or not having access to water buckets or automatic waterers.

Conclusions and Clinical Relevance—Analysis indicates that exposure to pasture predisposed mares to having MRLS-LTAs and stillborn foals. Methods for limiting exposure to pasture (keeping mares in stalls longer) during environmental conditions similar to those seen in 2001 should reduce the risk of mares having MRLS-LTAs. (J Am Vet Med Assoc 2003;222:199–209)

During the spring of 2001, there was an epidemic of abortions and stillbirths in mares in central Kentucky; these abortions and stillbirths were a component of mare reproductive loss syndrome (MRLS). Many of the abortions occurred in late gestation (ie, late-term abortions [LTAs]). The cause of MRLS remains unknown.

The objective of the study reported here was to compare data for mares that had MRLS-associated LTAs (MRLS-LTAs) with data for control mares that aborted for reasons other than MRLS or pregnant mares that did not abort, which would enable us to identify risk factors for mares having MRLS-LTAs. The term LTA was used to refer to abortions during late gestation as well as stillbirths.

Materials and Methods

Study population—Mares included in the study were selected on the basis of record review. Records from the University of Kentucky Livestock Disease Diagnostic Center (LDDC) were reviewed to identify aborted equine fetuses and stillborn foals that were submitted between January 1, 2001, and July 1, 2001. Initially, records were reviewed by 1 investigator (LRH) who assigned a global assessment score of the likelihood that a fetus was a MRLS-LTA. This assessment was made on the basis of the investigator's summary interpretation of the results of anamnesis, gross and microscopic pathologic findings, and microbiologic, toxicologic, and virologic testing. A fetus was considered a probable MRLS-LTA when the following criteria were met: assigned a global assessment score of being a MRLS-LTA by the investigator; >269 days of gestational age; and results of microbiologic, toxicologic, virologic, or other testing did not definitively identify a likely cause of abortion. A fetus was considered a definite MRLS-LTA when it met the 3 criteria for a probable MRLS-LTA and had at least 2 of the following additional elements: isolation of Streptococcus spp or Actinobacillus spp from fetal tissues, history of premature placental separation (ie, red-bag delivery), marked placental thickening or edema, placenta previa or infection of the umbilical cord, fibrinous pericardial effusion, colloidal distension of thyroid follicles, microscopic pulmonary lesions including squamous epithelial cells in alveoli and pulmonary parenchymal infiltration with leukocytes or monocytes-macrophages, or lack of any tissue lesions in the fetus. These criteria were developed by the authors in conjunction with veterinary pathologists at the LDDC. A fetus was also considered as a probable MRLS-LTA when it had 2 elements of the aforementioned list but did not have a global assessment score of being a MRLS-LTA as determined by the investigator.

Records of all fetuses submitted to the LDDC were reviewed to identify aborted fetuses >269 days of gestational age or fetuses that were stillborn and classified as a definite MRLS-LTA, probable MRLS-LTA, or other LTA (OLTA). Foals that died at ≥1 day of age (ie, born alive) were excluded. The following factors were recorded for each LTA: date of sub-
mission, LDDC admission number, gestational age, breed, sex, zip code, submitting clinic (whether it was either of 2 large equine clinics or another clinic), global assessment of MRLS-LTA (yes or no), pathologist, and whether any of the aforementioned specific pathologic findings were detected (eg, fibrinous pericardial effusion).

In the study reported here, data for case mares (ie, mares that had abortions deemed to be MRLS-LTAs) were compared with data for control mares (ie, mares that did not have MRLS-LTAs). The case group (MLTA group) comprised case mares from 2 large equine clinics as well as other MRLS-LTA mares that were identified at the farms of case mares by veterinarians who were not affiliated with either of the 2 equine clinics. Two groups of control mares were identified. The first control population (OLTA group) comprised mares that had aborted fetuses >209 days of age or had stillborn foals between January 1, 2001 and July 1, 2001 that were neither probable nor definite MRLS-LTA and were submitted by veterinarians from either of the 2 large equine clinics. Fetuses or stillborn foals that had a definitive diagnosis for abortion (eg, abortion attributable to herpesvirus infection) were eligible to be included in the OLTA group. The second control population (herd-mate controls; HERD group) was derived by, whenever possible, selecting mares from the same farm as MRLS-LTA mares; these were mares that gave birth to live foals during 2001 and were matched (+7 days) on the basis of breeding date with a mare that aborted. The rationale for selecting 2 control populations was that each control population would offset the inherent limitations of the other control group and that factors consistently identified as significantly associated with a mare being in the MLTA group, compared with both control groups, would have particular importance.

Data collection and analysis—For each case and control mare, information about the following general categories were recorded: name or farm identification number of the horse, signalment, breeding date and reproductive history, duration during which the mare resided in Kentucky, feeding practices, watering practices, stabling or housing practices, characteristics of pasture, pasture management practices, vaccination and deworming history, exposure to wildlife, and exposure to eastern tent caterpillars.

The association between categoric variables (eg, breed and MLTA group) was summarized by use of contingency tables for categoric data and medians and ranges for continuous data. These summaries neglected the correlation among observations from the same farm (ie, multiple case mares submitted from the same farm and the same farm contributing to the MLTA, OLTA, and HERD groups) and were used strictly for descriptive purposes. To account for the correlation among observations when examining the association between a given covariate and outcome category of abortion (eg, MLTA vs OLTA groups), an extension of generalized linear modeling referred to as generalized estimating equations was used for analysis. For purposes of multivariate analysis, some continuous covariates were categorized on the basis of results of exploratory data analysis (eg, use of the median value of the MLTA group as a cutoff). Data were analyzed by use of commercial and downloadable computer software.

A value of $P \leq 0.05$ was used to designate significance for all analyses. Methods for matched analysis (eg, conditional logistic regression) were not used for the comparison of the MLTA and HERD groups, because herd-mate control horses were not identified for all MLTA horses; however, the generalized estimating equations accounted for the correlation among observations for mares from the same farm.

The association between a given exposure (eg, being in pasture after midnight) and disease was expressed as the odds ratio (OR), which was derived from analyses that used generalized estimating equations. The 95% confidence intervals were calculated for these ORs by use of robust estimates of standard errors derived from generalized estimating equation analysis.

Results

Study population—Review of records of the LDDC revealed that 168 MRLS-LTAs were submitted by veterinarians from the 2 large equine clinics. We obtained data for 137 (82%) of these MRLS-LTA mares; owners of the remaining 31 MRLS-LTA mares declined to participate. Of the 137 MRLS-LTAs, 125 (91.2%) were classified as definite MRLS-LTAs, and the remaining 12 fetuses were classified as probable MRLS-LTAs.

We obtained data for 47 mares in the OLTA group (ie, non-MRLS-LTAs) and 98 mares in the HERD group (ie, mares that delivered a live foal from the same farms as the MLTA group and were matched on the basis of breeding date with mares that had MRLS-LTAs).

The 282 mares were from 62 farms. Each farm provided 1 to 24 mares (median, 3 mares). Sixteen farms had mares that were in both the MLTA and OLTA groups, 12 farms had mares that were in the OLTA group only, and 34 farms had mares that were in the MLTA group only.

Twelve counties were represented by mares included in the study. The counties contributing the most mares were Fayette (n = 88), Woodford (71), Bourbon (55), Scott (30), and Jessamine (27). Bourbon county seemed underrepresented among mares in the OLTA group (3 mares vs 27 in the MLTA group and 25 in the HERD group).

Comparison of MLTA and OLTA groups—Data for the MLTA group were compared with data for the OLTA group.

Dates of abortion and breeding—Mares in the MLTA group aborted between April 24 and May 26, 2001 (median, May 5). Mares in the OLTA group aborted between January 23 and May 14, 2001 (median, March 10). Breeding dates for mares in the MLTA group ranged from April 7 to August 29, 2000 (median, June 14). The MLTA group had earlier breeding dates (median, April 25, 2000; range, February 20 to June 30); however, the percentage of mares in the OLTA group bred before April 1, 2000 (median, 48%; range, 0 to 75%) was not significantly greater than that for mares in the MLTA group (median, 40%; range, 0 to 75%).

Age and parity—Age of mares in the MLTA group (median, 10 years; range, 4 to 28 years) was not significantly different from age of mares in the OLTA group (median, 10 years; range, 4 to 23 years). Similarly, there was not a significant difference in parity among mares in the MLTA (median, 4 foals; range, 1 to 17 foals) and OLTA (median, 4 foals; range, 1 to 13 foals) groups.

Breed—Breed of horses differed significantly among the
MLTA and OLTA groups (Table 1). Standardbreds were significantly (P = 0.007) over-represented among the MLTA group. These Standardbreds were from 3 farms.

Sex of fetus

We did not detect a significant difference in sex distribution of fetuses among MLTA and OLTA groups. For the MLTA group, 41% of fetuses were male, 47% were female, and sex was undetermined for 12%. For the OLTA group, 45% were male, 40% were female, and sex was undetermined for 15% of fetuses.

Microbial culture of fetal tissues

Tissues of all fetuses in the MLTA and OLTA groups were submitted for microbial culture. There were significantly more fetuses from which *Streptococcus* spp (P < 0.001) or *Actinobacillus* spp (P = 0.001) were isolated in the MLTA group (75/137 [54.7%] and 29/137 [21.2%], respectively) than in the OLTA group (74/47 [14.9%] and 1/47 [2.1%], respectively).

MRLS-associated conditions at the farm

One mare in the MLTA group was from a farm that

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**Table 1—Comparison of characteristics that differed significantly between 137 mares with late-term abortions (LTAs) associated with mare reproductive loss syndrome (MRLS; MLTA group) and 47 mares with LTAs attributed to other causes not associated with MRLS (OLTA group)**

<table>
<thead>
<tr>
<th>Variable</th>
<th>MLTA group</th>
<th>OLTA group</th>
<th>OR (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breed</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thoroughbred</td>
<td>86 (62.8%)</td>
<td>42 (89.4%)</td>
<td>1</td>
<td>NA</td>
</tr>
<tr>
<td>Standardbred</td>
<td>36 (26.3%)</td>
<td>4 (8.5%)</td>
<td>4.1 (1.3, 12.4)</td>
<td>0.007</td>
</tr>
<tr>
<td>Other</td>
<td>15 (10.9%)</td>
<td>1 (2.1%)</td>
<td>1 NA</td>
<td></td>
</tr>
<tr>
<td>Other MRLS-associated LTAs at the farm</td>
<td>121 (88.3%)</td>
<td>27 (57.4%)</td>
<td>5.3 (2.0, 13.9)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Median (range) amount of sweet feed fed daily (kg [lb])*</td>
<td>0 (0–7.3 [0–16])</td>
<td>3.6 (0–6.4 [0–14])</td>
<td>0.4 (0.2, &lt; 1.0)</td>
<td>0.034</td>
</tr>
<tr>
<td>Fed concentrate in pasture on ground during 4-week period prior to abortion</td>
<td>52 (65.8%)</td>
<td>5 (23.8%)</td>
<td>3.2 (1.3, 7.9)</td>
<td>0.007</td>
</tr>
<tr>
<td>Fed hay exclusively in pasture during 4-week period prior to abortion</td>
<td>41 (30.5%)</td>
<td>4 (8.5%)</td>
<td>4.2 (1.2, 14.0)</td>
<td>0.011</td>
</tr>
<tr>
<td>Water available in a bucket</td>
<td>98 (71.5%)</td>
<td>44 (93.0%)</td>
<td>0.1 (&lt; 0.1, 0.5)</td>
<td>0.001</td>
</tr>
<tr>
<td>Water available from automatic waterer</td>
<td>92 (67.2%)</td>
<td>43 (91.5%)</td>
<td>0.2 (&lt; 0.1, 0.5)</td>
<td>0.001</td>
</tr>
<tr>
<td>Median (range) number of hours in a stall during 4-week period prior to abortion</td>
<td>9 (0–24)</td>
<td>8 (0–24)</td>
<td>1.25 (0.8, 1.8)</td>
<td>0.001</td>
</tr>
<tr>
<td>Feed hay exclusively in pasture during 4-week period prior to abortion</td>
<td>12 (60.0%)</td>
<td>16 (80.0%)</td>
<td>0.4 (0.2, 0.8)</td>
<td>0.001</td>
</tr>
<tr>
<td>Water available in a trough</td>
<td>66 (48.2%)</td>
<td>9 (19.1%)</td>
<td>4.4 (1.8, 10.7)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Median (range) number of hours in a large paddock during 4-week period prior to abortion</td>
<td>9 (0–24)</td>
<td>8 (0–24)</td>
<td>1.25 (0.8, 1.8)</td>
<td>0.001</td>
</tr>
<tr>
<td>Pasture access after midnight during the 4-week period prior to abortion</td>
<td>82 (59.9%)</td>
<td>13 (27.6%)</td>
<td>3.8 (1.7, 8.3)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Mare vaccinated against rotavirus</td>
<td>44 (32.1%)</td>
<td>30 (63.8%)</td>
<td>0.3 (0.1, 0.6)</td>
<td>0.002</td>
</tr>
</tbody>
</table>

*Considered as a dichotomous variable of whether mares were fed ≥ 3.6 kg (8 lb) of sweet feed daily. †Odds ratios for 8-hour increments (ie, extent to which risk is estimated to decrease per 8 hours of additional time spent in a stall). ‡Odds ratios for 8-hour increments (ie, extent to which risk is estimated to increase per 8 hours of additional time spent in a large paddock). §Odds ratio for 10% increments (ie, extent to which risk increased for each 10% increase in dietary intake that was attributed to pasture).

OR = Odds ratios, which were derived by use of generalized estimating equations. 95% CI = 95% confidence interval. ND = Not determined. NA = Not applicable.
did not have any other mares (ie, no other horses at risk). Mares in the MLTA group were significantly \( P < 0.001 \) more likely than mares in the OLTA group to have a history of other MRLS-LTAs at the farm (Table 1).

**Breeding history and number of mares at the farm**

We did not detect significant differences between the MLTA and OLTA groups for the number of broodmares at the farm, number of mares that were transported to the farm to be bred, history of abortions during the preceding 5 years, duration during which the mare resided in Kentucky, and duration of residence on the farm.

**Feeding practices**

Only 3 mares were not fed any type of concentrate (all 3 were in the MLTA group). Although the proportion of mares in the MLTA group that were not fed sweet feed (72/137, 52.6%) was greater than that of the OLTA group (18/47, 38.3%), the values did not differ significantly. However, mares in the MLTA group were fed significantly \( P = 0.034 \) less sweet feed than mares in the OLTA group. For purposes of analysis, we considered a categoric variable of whether mares were fed \( \geq 3.6 \text{ kg (8 lb)} \) of sweet feed, which was the median amount fed to mares in the OLTA group (Table 1). There was not a significant difference in the proportion of mares in the MLTA and OLTA groups that were fed pellets or oats, nor in the amount of pellets or oats fed daily. Other concentrates (eg, corn) were fed too infrequently to enable us to make any inferences.

Although the proportion of mares in the MLTA group fed concentrate exclusively in pasture during the 4 weeks prior to abortion (39/137, 28.5%) was greater than that of the OLTA group (9/47, 19.1%), the difference was not significant. There were 40 mares in the MLTA group and 12 mares in the OLTA group fed concentrate partially in pasture and partially in stalls. Among the mares that were fed in pasture (either exclusively or partially), the MLTA group was significantly \( P = 0.007 \) more likely to have been fed concentrate on the ground (Table 1).

Most mares in the MLTA (111/137, 81.0%) and OLTA (42/47, 89.4%) groups were fed twice daily during the 4-week period prior to abortion. Only 9 mares in the MLTA group and 3 mares in the OLTA group had a change in diet during the 4-week period prior to abortion. The type of hay fed during the 4-week period prior to abortion or the source of hay (homegrown, grown in Kentucky, or grown outside Kentucky) did not differ between the OLTA and MLTA groups.

The location where hay was fed during the 4-week period prior to abortion differed significantly between the MLTA and OLTA groups. Of the mares in the MLTA group, 19 were fed hay exclusively in a stall, 41 were fed exclusively in pasture, and 77 were fed partially in a stall and partially at pasture; for mares in the OLTA group, the respective numbers were 7, 4, and 36. The MLTA group was significantly \( P = 0.011 \) more likely than the OLTA group to have been fed hay exclusively in pasture (Table 1). Hay fed to all mares was harvested almost exclusively during 2000. The groups did not differ significantly with regard to whether any dietary supplements, including salts, were fed.

**Watering practices**

For the 4-week period prior to abortion, the source of water (well, pond, or municipal water) and frequency with which waterers were cleaned did not differ significantly between MLTA and OLTA groups. Mares in the MLTA group were significantly \( P = 0.001 \) less likely than mares in the OLTA group to have water provided in a bucket or from an automatic waterer (Table 1). Conversely, the MLTA group was significantly \( P < 0.001 \) more likely than the OLTA group to have access to water from a trough.

**Stabling or housing practices**

During the 4-week period prior to abortion, only 1 mare in the MLTA group and 2 mares in the OLTA group did not have access to pasture for grazing. For each of the 4 weeks prior to abortion, the number of hours each horse was housed in a stall, maintained in a small paddock, and maintained in a large paddock was recorded. Mares in the MLTA group spent significantly \( P < 0.001 \) less time in a stall during each of the 4 weeks prior to abortion (Table 1). This difference was more pronounced for the third and fourth weeks prior to abortion.

For each of the 4 weeks prior to abortion, mares in the MLTA group spent significantly more time in small (\( < 0.405 \text{ hectares (\( < 1 \text{ acre} \))} \) paddocks (data not shown; median was 0 hours for both groups for each week). During the week prior to abortion, 20 mares in the MLTA group and 1 mare in the OLTA group spent time in a small paddock. Two weeks prior to abortion, 18 mares in the MLTA group and 1 mare in the OLTA group spent time in a small paddock. Three weeks prior to abortion, 13 mares in the MLTA group but none of the mares in the OLTA group spent time in a small paddock. Finally, 4 weeks prior to abortion, 12 mares in the MLTA group but none of the mares in the OLTA group spent time in a small paddock.

For each of the 4 weeks prior to abortion, mares in the MLTA group spent more time in large paddocks (\( > 0.405 \text{ hectares} \) than did mares in the OLTA group, and the amount of time was significantly different for the second, third, and fourth weeks prior to abortion (Table 1). It appeared that there was a pattern of greater time spent in large paddocks during the weeks furthest (ie, weeks 3 and 4) from abortion.

During the 4-week period prior to abortion, the percentage of each mare's daily intake estimated to be derived from pasture was significantly \( P < 0.001 \) greater for the MLTA group than for the OLTA group (Table 1). The MLTA group was significantly \( P < 0.001 \) more likely than the OLTA group to have access to pasture after midnight during the 4 weeks prior to abortion.

**Pasture characteristics**

The MLTA and OLTA groups did not differ significantly for any of several covariates, including access to pasture characterized as grass or grass and legumes during the 4-week period prior to abortion, the predominant type of roughage in pasture during the 4-week period prior to abortion, whether the amount of various grasses or legumes was larger than usual in the pasture where the mare resided during the 4-week period prior to abortion, whether an excess of white clover was observed in the pasture, and the amount of pasture used after midnight during the 4 weeks prior to abortion.
pasture where the mare resided during April and May of 2001 relative to April and May of 2000, whether hemlock was observed in the pasture where the mare resided during the 4-week period prior to abortion, whether pasture was described as lush during the 4-week period prior to abortion, history of fescue toxicosis at the farm, number of horses sharing the pasture, amount of land (hectares) in the pasture in which the mare was kept during the 4-week period prior to abortion, and total amount of land for the farm.

Pasture management

We did not detect significant differences between the MLTA and OLTA groups for several covariates, including whether pastures in which mares resided had previously been used for horses, whether pastures in which mares resided during the 4-week period prior to abortion had been rested from grazing by horses, whether the pasture in which the mare resided had been fertilized during the 4-week period prior to abortion, whether the pasture in which the mare resided during the 4-week period prior to abortion had been fertilized during the fall of 2000, type of fertilizer applied to pastures, whether lime had been applied to the pasture during 2000 or 2001, whether pastures had been mowed during the 4-week period prior to abortion, history of the pasture being mowed before the frost in mid-April of 2001, number of times the pasture had been mowed between January and May of 2001, whether manure was spread on the pasture in which the mare resided during the 4-week period prior to abortion, whether manure spread on the pasture had been composted prior to spreading, and whether herbicides or pesticides had been applied to the pasture during the 4 weeks prior to abortion. None of the pastures in which mares in the MLTA or OLTA groups were housed had pesticides or herbicides applied during the 4-week period prior to abortion.

Differences between the MLTA and OLTA groups were not identified for several variables, including whether there were cherry trees inside or overhanging the pasture in which the mare resided during the 4-week period prior to abortion, number of cherry trees inside or overhanging the pasture in which the mare resided during the 4-week period prior to abortion, whether there were cherry tree seedlings in the pasture in which the mare resided during the 4-week period prior to abortion, whether there were other fruit trees inside or overhanging the pasture during the 4-week period prior to abortion, whether deciduous trees in the pasture had been stripped of deciduous leaves, and whether pesticide was applied to trees in or around the pasture during the 4-week period prior to abortion.

Vaccinations and anthelmintics

The MLTA and OLTA groups did not differ with respect to history of administration of vaccines or anthelmintics, except that vaccination against rotavirus was significantly (P = 0.002) less likely among mares in the MLTA group than among mares in the OLTA group (Table 1).

Wildlife

The MLTA and OLTA groups did not differ significantly with regard to the frequency with which mice or rats, raccoons or skunks, coyotes or foxes, deer or elk, water fowl, bats, or opossums were observed on the premises or immediate vicinity during the 12-month period prior to interview of the owner or farm manager.

Eastern tent caterpillars

The concentration of Eastern tent caterpillars (hereafter referred to as caterpillars) at the farm during 2000 was characterized as heavy (blankets of caterpillars on fences, waterers, and other objects), moderate (many caterpillars in trees, some in pastures or barns), low (only a few caterpillars observed), or absent (Table 2). For purposes of statistical comparison, various categories were collapsed on the basis of statistical considerations (too few observations for a given category) or biological reasons (eg, moderate category more closely resembled the heavy category than the low category). After excluding those mares for which data were missing and combining the heavy and moderate categories, there was a significant (P = 0.043) difference in the estimated caterpillar burden of farms with mares in the MLTA or OLTA groups during 2000. Farms with mares in the MLTA group appeared to be more likely to have a caterpillar burden characterized as low than did farms with mares in the OLTA group. However, there was not a significant difference between MLTA and OLTA groups when comparisons were made of heavy or moderate caterpillar burden versus low caterpillar burden or absence of caterpillars observed.

The same categories of caterpillar burden were used to obtain data for 2001. Both MLTA and OLTA groups had heavier burdens of caterpillars during 2001 relative to 2000. There was not a significant difference between the MLTA and OLTA groups in the proportion of categories of caterpillar burden observed during 2001. When the moderate and low categories or the heavy and moderate categories were combined for purposes of comparison, there was not a significant difference between the MLTA and OLTA groups.

We did not detect significant differences between groups in the proportions for which caterpillars were observed in trees in or around the pasture of the affected

Table 2—Data regarding concentration of eastern tent caterpillars for mares that had MRLS-associated LTAs (MLTA group), LTAs attributed to causes other than MRLS (OLTA group), and mares selected from the same farms as the MLTA group but that did not abort and were matched on the basis of breeding date (HERD group)

<table>
<thead>
<tr>
<th>Variable</th>
<th>MLTA group</th>
<th>OLTA group</th>
<th>HERD group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Concentration of caterpillars observed during 2000</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heavy</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Moderate</td>
<td>31</td>
<td>17</td>
<td>22</td>
</tr>
<tr>
<td>Low</td>
<td>80</td>
<td>20</td>
<td>51</td>
</tr>
<tr>
<td>None</td>
<td>9</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>Did not recall</td>
<td>16</td>
<td>3</td>
<td>18</td>
</tr>
<tr>
<td>Concentration of caterpillars observed during 2001</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heavy</td>
<td>123</td>
<td>39</td>
<td>89</td>
</tr>
<tr>
<td>Moderate</td>
<td>8</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>Low</td>
<td>2</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>None</td>
<td>0</td>
<td>0</td>
<td>0</td>
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</tbody>
</table>
and 3 foals for which sex was not known. For the MLTA group, 56 fetuses were male, 65 fetuses were female, and 16 fetuses were of unknown sex. For the HERD group, 59.2% of foals born were male, whereas 71.4% of foals born to mares in the MLTA group were male. The difference between the MLTA (86/137, 62.8%) and HERD (59/84, 70.2%) groups was not statistically significant. Although the proportion of Standardbreds in the MLTA group was smaller for the MLTA group (36/137, 26.3%) than the HERD group (58/98, 60.2%) groups, the proportion of Standardbreds was similar between the MLTA (86/137, 62.8%) and HERD (58/98, 59.2%) groups. All of the variables examined remained significantly associated with being in the MLTA group except for feeding (as would be anticipated from our study design), which remained significantly associated with being in the MLTA group, analyses were repeated for each of several variables and included a term for Standardbred breed (ie, each association was examined while adjusting for the effects of Standardbred breed). All of the variables examined remained significantly associated with being in the MLTA group except for feeding ≥ 3.6 kg of sweet feed. Even though the significance was changed for this variable, neither the magnitude nor the direction of the observed, unadjusted association was changed.

Comparison of MLTA and HERD groups—Data for the MLTA group were compared with data for the HERD group.

Dates of breeding and abortion

Mares in the MLTA group aborted between April 24 and May 26, 2001 (median, May 5). Mares in the HERD group foaled between April 18 and June 14, 2001 (median, May 12). Breeding dates for mares in the MLTA group ranged from April 7 to August 29, 2000 (median, June 14), and were similar to those for the HERD group (as would be anticipated from our study design), which ranged from April 8 to July 13, 2000 (median, June 8).

Age and parity

Age of mares in the MLTA group (median, 10 years; range, 4 to 28 years) was not significantly different from mares in the HERD group (median, 10 years; range, 4 to 23 years). Similarly, there was not a significant difference in parity among mares in the MLTA (median, 4 foals; range, 1 to 17 foals) and HERD (median, 4 foals; range, 1 to 18 foals) groups.

Breed

The proportion of Thoroughbreds was similar between the MLTA (86/137, 62.8%) and HERD (58/98, 59.2%) groups. Although the proportion of Standardbreds was smaller for the MLTA group (36/137, 26.3%) than the HERD group (36/98, 36.7%), the proportions did not differ significantly between groups.

Sex of fetus

Sex distribution of aborted fetuses or stillborn foals of mares in the MLTA group did not differ significantly from that of foals born to mares in the HERD group. For the MLTA group, 56 fetuses were male, 65 fetuses were female, and 16 fetuses were of unknown sex. For the HERD group, there were 45 male foals, 30 female foals, and 3 foals for which sex was not known.

Breeding history and number of mares at the farm

The number of mares at farms for the MLTA group was significantly (P = 0.006) less than that of the HERD group (Table 1). This indicated that the HERD group was preferentially selected from larger farms.

We did not detect significant differences between mares in the MLTA and HERD groups for several covariates, including history of previous abortion during the preceding 5 years, number of mares that were transported to the farm to be bred, proportion of mares bred before April 1, duration during which the mare resided in Kentucky, and duration of residence at the farm.

Feeding practices

All mares except 3 in the MLTA group were fed concentrate. There was not a significant difference between MLTA and HERD groups in the type or amount of concentrate or hay fed, where concentrate was fed (stall vs pasture), whether concentrate fed in pasture was offered on the ground or above the ground, frequency of daily feeding, or history of a recent change in diet. For the HERD group, the most common change in diet was addition of a commercial mycotoxin binder (alone [7 mares] or in combination with dexamethasone [5]); only 1 mare in the MLTA group was fed mycotoxin binder, and the proportions of mares in the HERD (12/98, 12.2%) and MLTA (1/137, 0.7%) groups fed mycotoxin binder differed significantly (P < 0.001). Twelve of the 13 mares fed mycotoxin binder were from 2 farms. These data could not be analyzed by use of generalized estimating equations because of complete separation (ie, a cell that contained a value of 0) after accounting for farm effects.

The MLTA group was significantly more likely than the HERD group to have been fed alfalfa hay (Table 3). There was not a significant difference between MLTA and HERD groups with regard to the proportions fed hay that was homegrown, grown elsewhere in Kentucky, or grown outside of Kentucky during the 4-week period prior to abortion or foaling.

The proportion of mares in the MLTA group fed hay only in the pasture during the 4-week period prior to abortion (41/137, 29.9%) was not significantly different from that of mares in the HERD group (24/98; 24.5%) during the 4-week period prior to foaling. Except for 1 mare in the MLTA group, all hay fed to mares was harvested in 2000. There was not a significant difference between groups in the proportion of mares fed dietary supplements (including salts) during the 4 weeks prior to abortion or foaling.

Watering practices

For the 4-week period prior to abortion (MLTA group) or foaling (HERD group), source of water (well, pond, or municipal water), types of waterers provided (buckets, troughs, automatic waterers, or other sources), and frequency with which waterers were cleaned did not differ significantly between MLTA and HERD groups.

Stabling or housing practices

The estimated number of hours each day that each mare was kept in a stall, small paddock, or large paddock was determined for each of the 4 weeks prior to
arbitration (MLTA group) or foaling (HERD group). Compared with the HERD group, mares in the MLTA group spent significantly fewer hours in a stall and significantly more hours in large paddocks during the 4-week period prior to abortion (Table 3). There appeared to be a pattern of increasing difference for the number of hours in each situation with increasing time from the date of foaling or abortion.

All but 3 mares (1 in the MLTA group and 2 in the HERD group) had access to pasture for grazing during the 4-week period prior to abortion or foaling. There was a significant difference between MLTA and HERD groups in which mares were grazing on pasture after midnight during the 4-week period prior to abortion (HERD group) or foaling (MLTA group).

Pasture characteristics

We did not detect significant differences between the MLTA and HERD groups for several covariates, including access to grazing grass or a mixture of grass and legumes during the 4-week period prior to abortion or foaling, history of amounts of white clover that were larger than usual in the mare’s pasture during the 4-week period prior to abortion or foaling, whether hemlock was observed in the pasture where the mare resided during the 4-week period prior to abortion or foaling, pastures described as being lush during the 4-week period prior to abortion, number of horses sharing the pasture, or total amount of land for the farm. Pastures of the MLTA group were significantly (P < 0.001) smaller than those of the HERD group (Table 3).

Total amount of land for farms for the MLTA group was significantly (P = 0.004) less than that of farms for the HERD group.

Pasture management

Only 3 mares (all in the MLTA group) were in pastures that had not previously been used as pasture for horses. We did not detect significant differences between the MLTA and HERD groups for several covariates, including whether pastures had been rested from grazing by horses prior to use during 2001, whether pastures had been fertilized during the 4-week period prior to abortion or foaling, whether pastures had been fertilized during 2000, whether lime had been applied to pastures during the fall of 2000 or spring of 2001, whether pastures had been mowed during the 4-week period prior to onset of abortion or foaling, whether pastures had been mowed before the frost in mid-April of 2001, number of times that pastures had been mowed during the 4-week period prior to abortion. Manure that was spread had been composted for 4 mares in the MLTA group and 1 mare in the HERD group. Neither herbicides nor pesticides had been applied to any of the pastures in which the mares of either group resided during the 4-week period prior to abortion or foaling. We did not detect significant differences between the MLTA and HERD groups for several other covariates, including whether there were cherry trees inside or overhanging the pasture during the 4-week period prior to abortion or foaling, number of cherry trees inside or overhanging pastures, whether there were

<table>
<thead>
<tr>
<th>Variable</th>
<th>MLTA</th>
<th>HERD</th>
<th>OR (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median (range) number of mares at the farm</td>
<td>60 (1–292)</td>
<td>75 (12–292)</td>
<td>&lt; 1* (0.9, &lt; 1)</td>
<td>0.006</td>
</tr>
<tr>
<td>Median (range) amount of each pasture that mare was kept in during the 4-week period prior to abortion (MLTA group) or foaling (HERD group) (hectares [acres])</td>
<td>37.0 (15) (2.5–170.3) (1–69)</td>
<td>49.4 (20) (2.5–170.3) (1–69)</td>
<td>0.87 (0.7, 0.9)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Median (range) amount of land for farm (hectares [acres])</td>
<td>121 (300) (0.4–648) [1–1,600]</td>
<td>165 (407) (12–648) [30–1, 600]</td>
<td>0.91 (&lt; 0.9, &lt; 1)</td>
<td>0.004</td>
</tr>
<tr>
<td>Fed alfalfa hay</td>
<td>Yes</td>
<td>33 (24.8%)</td>
<td>13 (13.5%)</td>
<td>2.1 (1.0, 4.4)</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>100 (75.2%)</td>
<td>83 (86.5%)</td>
<td>1</td>
</tr>
<tr>
<td>Median (range) number of hours in a stall during 4-week period prior to abortion (MLTA group) or foaling (HERD group)</td>
<td>1 week prior</td>
<td>12 (0–24)</td>
<td>15 (0–24)</td>
<td>0.55 (0.3, 0.8)</td>
</tr>
<tr>
<td></td>
<td>2 weeks prior</td>
<td>11 (0–24)</td>
<td>15 (0–24)</td>
<td>0.66 (0.4, 0.9)</td>
</tr>
<tr>
<td></td>
<td>3 weeks prior</td>
<td>0 (0–18)</td>
<td>13 (0–24)</td>
<td>0.75 (0.4, 0.9)</td>
</tr>
<tr>
<td></td>
<td>4 weeks prior</td>
<td>0 (0–18)</td>
<td>12 (0–24)</td>
<td>0.75 (0.5, &lt; 1.0)</td>
</tr>
<tr>
<td>Median (range) number of hours in a large paddock during 4-week period prior to abortion (MLTA group) or foaling (HERD group)</td>
<td>1 week prior</td>
<td>9 (0–24)</td>
<td>9 (0–24)</td>
<td>1.68 (1.2, 2.3)</td>
</tr>
<tr>
<td></td>
<td>2 weeks prior</td>
<td>10 (0–24)</td>
<td>9 (0–24)</td>
<td>1.45 (1.1, 1.9)</td>
</tr>
<tr>
<td></td>
<td>3 weeks prior</td>
<td>16 (0–24)</td>
<td>9 (0–24)</td>
<td>1.55 (1.1, 2.0)</td>
</tr>
<tr>
<td></td>
<td>4 weeks prior</td>
<td>18 (0–24)</td>
<td>9 (0–24)</td>
<td>1.41 (1.1, 1.9)</td>
</tr>
<tr>
<td>Ivermectin administered during 4-week period prior to abortion (MLTA group) or foaling (HERD group)</td>
<td>Yes</td>
<td>42 (31.8%)</td>
<td>12 (12.9%)</td>
<td>3.2 (1.8, 5.4)</td>
</tr>
<tr>
<td></td>
<td>No</td>
<td>90 (68.2%)</td>
<td>81 (86.5%)</td>
<td>1</td>
</tr>
</tbody>
</table>

*Odds ratio per 10 mares. †Odds ratio per 10 acres. ‡Odds ratio per 100 acres. §Odds ratio per 8 hours.
cherries outside of the pastures, whether there were cherry tree seedlings inside the pasture, whether there were other fruit trees inside or overhanging the pastures, whether there were other fruit trees outside the pastures, and whether deciduous trees around the pastures were stripped of leaves during the spring of 2001. Eight mares in each of the 2 groups were in pastures for which trees in or near the pasture were treated with pesticides during the 4-week period prior to abortion or foaling.

Vaccinations and anthelmintics administered
We did not detect significant differences between MLTA and HERD groups with regard to administration of vaccines or anthelmintics, except that mares in the MLTA group were significantly more likely than mares in the HERD group to have received ivermectin during the 4-week period prior to abortion or foaling (Table 3).

Wildlife
We did not detect significant differences between the MLTA and HERD groups with respect to any type of wildlife exposure.

Eastern tent caterpillars
We did not detect significant differences between groups with respect to the estimated caterpillar burden during 2000. Both MLTA and HERD groups had heavier burdens of caterpillars during 2001 relative to 2000 (Table 2). There was not a significant difference between the MLTA and HERD groups in the caterpillar burden observed during 2001. There was not a significant difference between the 2 groups for whether caterpillars were observed in trees in or around the pasture of the affected mare or observed in the pasture where the mare aborted or foaled as well as in the estimated percentage of pastures at the farm that had evidence of caterpillars.

Multivariate analysis
Because several variables related to farm size were significantly associated with being in the MLTA group, analyses were repeated for each of those variables associated with being in the MLTA group (other than those related to farm size) that included a term for farm size (ie, each association was examined while adjusting for the effects of farm size). All variables, except for feeding alfalfa hay, remained significantly associated with a mare being in the MLTA group. Multivariate models also were fit by use of individual variables pertaining to duration of being at pasture or being in a stall (eg, number of hours spent in a large paddock during the 4-week period prior to abortion or foaling) and feeding of alfalfa hay. Of the 8 multivariate models evaluated, in only 1 did alfalfa hay remain significantly associated with a mare being in the MLTA group.

Discussion
The objective of the study reported here was to compare mares that had MRLS-LTAs with mares that did not have MRLS-LTAs (ie, control horses) to identify factors that increased the risk of mares having MRLS-LTAs. Identifying control populations for case-control studies is challenging, because there are limitations associated with all control groups that are identified. One method to overcome the limitations of any single control group is to select multiple control groups for comparison. In this study, 2 groups of control horses were selected (mares that had LTAs other than MRLS-LTAs during 2001 [OLTA group] and mares from the same farm as the MLTA group but that delivered live foals in 2001 and were matched on the basis of breeding date with the MLTA mares [HERD group]). The OLTA group was selected to identify factors that distinguished MRLS-LTAs from other LTAs. The HERD group was selected in an attempt to identify factors predisposing mares to being in the MLTA group within each farm (eg, factors that distinguished affected mares from unaffected mares with similar breeding dates at farms that had the problem). This study must be interpreted with consideration of results of comparison of the MLTA group with each of the control groups and with consideration of the strengths and limitations of each control group.

Analysis of the results of the study indicated that exposure to pasture was the most important risk factor for MRLS-LTAs. A number of factors reflecting exposure to pasture were significantly associated with a mare being in the MLTA group. Comparisons with both control groups indicated that MRLS-affected mares spent significantly less time in a stall and more time at pasture, particularly during the period 3 and 4 weeks prior to abortion. Given that the median abortion date for the MLTA group was May 5, 2001, these data are consistent with exposure in a pasture to a causal agent during the second or third week in April of 2001 and indicate the need for additional temporal studies of this epidemic.

Because mares in the OLTA group aborted significantly earlier in the year (median abortion date, March 10), this group may have had less exposure to pasture because of seasonal patterns in turnout time (ie, less exposure to pasture during colder weather); however, the consistency of the association with time at pasture, using control mares from the same farms that were closely matched for breeding date (HERD group), indicated that the association with increased time in pasture was not explained by seasonal differences between case and control mares. This association with pasture also did not seem to be confounded by stage of gestation (ie, mares at differing stages of gestation had differing exposure to pasture), because the HERD group was closely matched with the MLTA group on the basis of breeding date.

For the comparison of MLTA and OLTA groups, a number of other variables presumably related to being in pasture were significantly associated with increased risk of a mare being in the MLTA group. Among mares fed in pastures, those fed concentrate on the ground during the 4-week period prior to abortion were at increased risk of having a MRLS-LTA. This observation is consistent with the hypothesis that feeding on the ground increased the opportunity to ingest a toxic agent in or on the pasture grass; however, it may simply be a marker for some other management practice that increased the risk of a mare having a MRLS-LTA. The observation that mares in the MLTA groups were
fed less sweet feed may indicate that these mares were allowed to have greater dietary intake from roughage and pasture. Indeed, the median estimated percentage of daily dietary intake derived from pasture was significantly greater for the MLTA group than for the OLTA group. These differences may, however, reflect differences in some other management practice for mares bred earlier in the year (OLTA group), compared with those bred later in the year (MLTA group). Neither feeding concentrate on the ground nor the amount of sweet feed fed during the 4-week period prior to abortion or foaling differed between the MLTA and HERD groups; however, this may have been attributable to homogeneity of feeding practices within farms (ie, farms may have fed mares at similar stages of gestation in a similar manner).

The MLTA group was significantly more likely than the OLTA group to have been fed hay exclusively in pasture during the 4-week period prior to abortion. This finding likely reflected increased time in pasture for the MLTA group relative to the OLTA group. However, it may also have increased the opportunity for ingestion of a pasture-associated toxin when the hay was fed on the ground. Access to pasture after midnight during the 4-week period prior to abortion was significantly greater for the MLTA group than the OLTA group. Season or ambient temperature may have influenced this finding, because it was not observed when the MLTA group was compared with the HERD group.

The MLTA group was significantly more likely to have been fed alfalfa hay than was the HERD group. This effect may have been attributable to confounding (ie, the association of feeding alfalfa hay with another exposure causally associated with MRLS-LTAs). For example, duration of time at pasture was slightly greater for horses fed alfalfa hay; however, the association of feeding alfalfa hay was not consistently significant after adjusting for the duration of time at pasture or in a stall.

Water buckets and automatic waterers are often located in stalls, whereas water troughs are often placed in pastures. The observation that mares in the MLTA group were significantly more likely than mares in the OLTA group to have troughs as their water source, and significantly less likely than the OLTA group to have water available in buckets or from automatic waterers, was most probably explained by an increase in the amount of time at pasture for the MLTA group. Again, failure to identify similar associations with the HERD group may have been a reflection of similarity in management; alternatively, the analysis may have required refinement to reflect practices during each of the weeks prior to abortion (as indicated by results for the questions regarding duration of time spent in paddocks or stalls) rather than for the entire 4-week period prior to abortion.

The association of abortion with exposure to pasture is consistent with the hypothesis of a toxic agent in or on plants that were in or around the pasture as the causal agent of abortion. A number of plants (including grasses) have been associated with abortion in cattle and sheep. Few reports, however, describe plants that are fetotoxic in horses. Sudan grass and sorghum are considered toxic to equine fetuses. Various mycotoxins have been associated with abortion in food-producing animals, including zearalenone and aflatoxin. To the authors' knowledge, mycotoxins have not been reported as a cause of abortion in horses.

Because of the putative association of MRLS-LTs with events during mid-April of 2001, it was important that there be relatively similar proportions of mares in the MLTA and OLTA groups that may have been exposed during late gestation. The proportion of mares bred before April 1, 2000, was determined in an attempt to assess this. There was not a significant difference between the MLTA group and the OLTA or HERD groups in the percentage of mares bred before April 1, 2000.

A number of variables associated with size of the farm (number of mares at the farm, total amount of land, and amount of land for each pasture in which mares were kept) differed between the MLTA and HERD groups. This finding was attributed to the fact that it was easier to identify breeding-date matched control mares from larger farms with more mares. After adjusting for farm size, however, the association of MRLS-LTAs with duration of time in a stall or at pasture remained significant.

The MLTA group was significantly less likely than the HERD group to have recently had a commercially available mycotoxin binder added to the feed. Although these results could suggest a protective effect for these binders (and thereby implicate mycotoxins as the cause of MRLS-LTAs), these data must be interpreted with considerable caution. First, only a small number of mares (1 mare in the MLTA group and 12 mares in the HERD group) had this binder added to their feed during the 4-week period prior to abortion or foaling. More importantly, a limitation of retrospective data collection for case-control studies is that the temporal association of the event and the exposure often cannot be ascertained. It is possible, if not probable, that the mycotoxin binders were added to feed at farms that had mares with MRLS-LTAs after the MRLS-LTA events occurred. Thus, addition of the mycotoxin binder may have been an effect rather than a cause of the observed association. Moreover, the question posed only pertained to horses with a change in diet during the 4-week period prior to abortion. It is unclear as to the extent MLTA and HERD groups were being fed this binder prior to the specified time period. For these reasons, the apparent protective effect of the mycotoxin binder must be interpreted with caution. Additional corroborating evidence would be needed prior to recommending feeding such binders as a protective strategy.

The association of ivermectin administration with increased risk of a mare having a MRLS-LTA was difficult to explain. It is possible that this anthelmintic had some synergism with the causal agents or agents of MRLS-LTAs. Alternatively, farms that administered ivermectin to mares may have contributed more mares to the MLTA group than to the HERD group. Finally, this observation might have occurred by chance alone;
it was not corroborated by comparison of the MLTA and OLTA groups.

The significant association of MRLS with vaccination against rotavirus was difficult to explain. It likely reflected some other difference in health management practices or seasonal distribution of foaling between the farms for mares of the MLTA and OLTA groups. Alternatively, farms that had outbreaks of rotavirus infections may have been protected from MRLS-LTAs for reasons other than management. This observation must be interpreted cautiously because of the lack of additional biological or epidemiologic evidence that would support an association between vaccination against rotavirus and MRLS.

Exposure to cherry trees, eastern tent caterpillars, or both have been implicated as causes of MRLS.1 Analysis of results of this study indicated that there appeared to be an increase in exposure to caterpillars in 2001 relative to 2000. We did not find any evidence implicating cherry trees or caterpillars in the analyses for the study reported here, although multiple qualitative and semi-quantitative questions directed toward capturing data about these factors were used. One possible explanation for these discrepant results regarding cherry trees and caterpillars is that our horse-level studies may have been biased by obtaining data from case mares (MLTA group) and control mares from the same farm (HERD group) or from farms that may have had both MRLS-LTAs and other MRLS-related problems (OLTA group). With regard to the latter, the MLTA group was significantly more likely to have come from farms that had MRLS-LTAs than were mares in the OLTA group. If the association of MRLS-LTAs with cherry trees and caterpillars were strong, then we would have expected to identify a significant association at the horse level by use of same-farm controls (HERD group comparison), much as we saw an association with time at pasture in the comparison of the MLTA and OLTA groups and the MLTA and HERD groups. Another possible explanation for failure to identify significant associations is that questions to elicit data may not have accurately reflected exposures, because there are various species of tent caterpillars and cherry trees, such that a species-specific exposure may not have been identified in our study.

Another possible explanation for this discrepancy is that the previous study examined risk factors for farms that had heavy losses of early-term abortions (early fetal losses), whereas the study reported here evaluated LTAs. Because management of mares in late gestation likely differs from that of mares in early gestation, it is possible that factors influenced by management that were significantly associated with LTAs may not be associated with early-term abortion. Exposure of mares to caterpillars likely occurred in pasture; caterpillar burden may have been an indicator of pasture exposure in mares that had early-term abortion associated with MRLS. Alternatively, strong association of MRLS-LTAs with exposure to pasture may have reflected exposure to caterpillar-contaminated pastures.

Because farms contributed multiple cases, and because there were 16 farms that contributed mares to both the MLTA and OLTA groups, we used generalized estimating equations to account for the correlation among observations from farms. For the comparison of MLTA and OLTA groups, we also conducted a separate analysis excluding data from the 16 farms that contributed to both the MLTA and OLTA groups (ie, we used data from farms that contributed mares only to the OLTA group or only to the MLTA group). Results of this analysis (data not shown) yielded essentially identical results to those for the analysis that included these observations and that was conducted by use of generalized estimating equations.

The definition of a case of MRLS-LTA was made on the basis of review of records of the LDDC by the authors and the experiences and recommendations of other pathologists at the LDDC. To our knowledge, there have not been any reports regarding pathology-based characterization of MRLS-LTAs. Because a pathology-based definition of MRLS-LTAs (other than that proposed in the study reported here) was lacking, it is possible that some cases of MRLS-LTA and other LTAs were misclassified. The net effect of this misclassification would have been to bias our results toward unity (ie, away from finding significant differences between groups). Use of a control population of mares that did not abort helped to limit the impact of potential misclassification. Most of the cases of MRLS-LTA were categorized as definite; results of analyses were similar when the 12 MRLS-LTAs categorized as probable were excluded from analysis.

The finding that tissues from fetuses classified as MRLS-LTA were significantly more likely than tissues from other LTAs to have yielded positive results for microbiologic culture of Streptococcus spp or Actinobacillus spp was expected, because these were criteria for case definition. These microbiologic findings were used as criteria for case definition on the basis of the experiences and recommendations of pathologists at the LDDC. Whether these agents were pathogens, opportunists, or contaminants merits further investigation.

The Streptococcus isolates were identified as α-hemolytic streptococci. Such α-hemolytic streptococcal organisms can be recovered from the vagina and uterus of healthy mares.18 These organisms may have been contaminants acquired from the mare’s reproductive tract during expulsion of the fetus. Although streptococcal organisms can cause abortion in mares,17,18 α-hemolytic streptococcal organisms are rarely associated with abortion or placentalitis in mares.19,20 In rhesus monkeys, α-hemolytic streptococci have been associated with abortion attributed to ascending infection of commensal vaginal organisms.21 In humans, β-hemolytic streptococci have been associated with mid- and late-term abortions.22 These organisms have been associated with placentalitis and pneumonia, lesions that were commonly observed among the aborted fetuses in the mares of our report. Actinobacillus spp are rarely isolated from the reproductive tract of healthy mares,23 equine placental tissues,24 or aborted equine fetuses.25 Epidemiologic features of the MRLS epidemic, however, are not consistent with a transmissible infectious agent. Aborted fetuses were submitted from multiple counties during
a relatively short period. Although such a distribution of cases in time and place might occur with a community-wide point-source infection, it is not likely that an infectious agent was the cause.

The most important result of the study reported here was that pasture exposure predisposed mares to having MRLS-LTAs. Our findings indicated that methods for limiting exposure to pasture (keeping mares in stalls longer and feeding hay and concentrate above ground) may be used to prevent recurrence of the problem during environmental conditions similar to those that occurred during the spring of 2001 in central Kentucky. Because these findings apply to the unusual environmental conditions that occurred during the spring of 2001 in central Kentucky, restricting access of mares to pasture during more typical environmental conditions likely would not be necessary.

A cause of abortion usually can be identified during pathologic examination of equine fetuses. The cause of the MRLS epidemic, however, remains unknown.

References