Equine leucoencephalomalacia (ELEM) is a highly fatal disease affecting horses and other Equidae. The disease is most often characterized by an acute and severe neurologic disorder but is also reported to produce a hepatic syndrome manifested by icterus, generalized systemic hemorrhaging and edema (9).

ELEM has been recognized and reported in the United States since the latter part of the nineteenth century. Many names have been used in the literature to describe this condition including blind staggerers, cerebritis, leucoencephalitis, encephalomyelitis, cerebrospinal meningitis, foraging disease, corn stalk disease, and moldy corn poisoning. In the early 1900's an outbreak of an acute, fatal neurologic disease was described in horses fed moldy fodder (3). A disease with similar clinical signs and lesions was later experimentally produced in horses fed moldy corn (4). An estimated 5000 horses died in central Illinois during the late fall and winter of 1934-35 from a disease referred to as "cornstalk disease" (6). The signs reported were indicative of a neurologic disorder. Brain suspensions and filtrates from many of the field cases were inoculated into laboratory animals and horses to test for viruses of eastern and western encephalomyelitis. Test results indicated that the encephalomyelitis which occurred was not associated with either of these viruses. In the late 1950's ELEM was again experimentally produced in horses by feeding moldy corn obtained from natural occurring outbreaks in Iowa (2,10,11). No virus was demonstrated in the brains of the affected animals nor did laboratory animal inoculations of brain suspensions indicate the presence of viral agents. Cultures of brain and other organs were also negative for bacterial pathogens.

Numerous outbreaks of encephalomalacia have been reported in Egyptian Equidae along the Nile Delta. Serologic tests of many equine sera obtained from horses within the affected areas failed to demonstrate antibodies to the eastern, western, St. Louis and Japanese B encephalitis viruses. Attempts to isolate viral, bacterial, and parasitic agents from affected animals were also futile. A disease which produced signs and lesions characteristic of ELEM was thought to be of fungal origin and numerous fungi were isolated from suspected feeds but a specific species could not be incriminated. Several species of fungi were isolated from the Egyptian corn. Each fungal isolate was grown at room temperature on autoclaved kernels of American corn and fed to American and Egyptian donkeys. Only the corn infected with the fungus Fusarium moniliforme Sheldon produced clinical signs and lesions characteristic of ELEM (15).

An isolate of Fusarium moniliforme Sheldon was obtained from maize suspected of causing ELEM in South Africa. Three horses and three donkeys were dosed by stomach tube with autoclaved corn fermented with this isolate of Fusarium. Two horses and one donkey died showing clinical signs of subcutaneous edema and icterus. Histopathologic findings included a centrilobular hepatic fibroplasia, proliferation of bile ducts and fatty infiltration of hepatocytes. No gross brain lesions were seen and only small perivascular hemorrhages in the subcortical white matter were reported on histopathologic examination (8). This same Fusarium isolate was again cultured on corn and administered by stomach tube to two horses. Postmortem and histopathologic findings of one horse again revealed a hepatotoxicosis characterized by extensive centrilobular fibroplasia. No lesions were seen in the brain. The other horse had brain lesions characteristic of ELEM but did not have any hepatic lesions. The hepatic syndrome was induced by feeding 0.67 - 1.94 kg culture material each day for 11-21 days, whereas, the neurologic syndrome was induced by feeding 0.33 - 0.41 kg/day for 90-144 days (9).

In the United States ELEM has been a seasonal disease usually occurring from late fall through early spring (2,4,6,10). The clinical signs reported in the field cases of ELEM were variable but usually included a severe central nervous system (CNS) disorder. The first signs were usually intermittent anorexia, depression, and anemia. In some cases the lower lip drooped and the head was held in a lowered position. Impaired foodprehension and mastication were reported in some field cases. Incoordination, aimless walking or walking in a circle, blindness, and head pressing have
all been commonly reported. Signs of severe CNS derangement occurred abruptly with extreme unexplainable agitation, hyperexcitability, profuse sweating, and delirium. Terminal signs were lateral recumbency and clonic-tonic seizures with death occurring 1-12 hours after onset. The clinical signs reported in the hepatotoxic syndrome where neurologic deficits were not apparent, included swelling of the lips and nose, a lowered head, reluctance to move, severe icterus, petechial hemorrhages in the conjunctival, abdominal breathing, cyanosis, and death within 4 hours.

The gross neural lesions of ELEM are frequently described as severe and extensive, although there are reports of death with no lesions present. The most striking feature, considered pathognomonic by some workers, was one or more liquefactive necrotic lesions in the subcortical white matter of the cerebral hemispheres. These lesions varied in size from microscopic to several cm. The necrotic lesions occurred randomly within the white matter and some were bilateral. The white matter within the areas of necrosis was edematous and structureless. The glia cells showed pyknotic nuclei and eosinophilic cytoplasm. The area bordering the malacic lesions was described as having rarefication of the white matter, perivascular hemorrhages, and satellitosis. Necrosis was not limited to subcortical white matter. Malacic areas in the cerebral cortex and focci of necrosis in the gray matter of the spinal cord have been reported (9,7).

Other lesions reported were diffuse centriflobular hepatic fibrosis, fatty vacuolization of hepatocytes, bile duct proliferation, and hemorrhagic enteritis (7,8,9).

The characteristic brain lesions of ELEM have not been reported or reproduced in any other animal species. Rabbits, rats, guinea pigs, hamsters, mice, goats, sheep, pigs, and monkeys have all been dosed with pure cultures of F. moniliforme and no deaths or brain lesions were produced (12,15).

F. moniliforme is a common mold on corn. Infected corn kernels are recognized by a pink to reddish brown color. The fungus has been reported to grow from infected to healthy kernels after harvesting if moisture content was above 15%. Numerous mycotoxins have been isolated from F. moniliforme but very little information is known about their toxicity. Moniliformin was isolated from F. moniliforme Sheldon-infected corn. Purified toxin was monitored by a cockerel bioassay. The oral median lethal dose (LD₅₀) was 4.0 mg/kg body weight. Gross and histopathologic lesions in the birds that lived more than two hours after dosing included ascites, edema of the mesenteries, small hemorrhages in the proventriculus, gizzard, small and large intestines, and skin. Birds that died within two hours of dosing had no significant lesions (5). Other metabolites produced by F. moniliforme include zearalenone, malonic acid, fusariocin A, fusicarin acid, gibberellins, and kaurane diterpenoids (5,8).

**MATERIALS AND METHODS**

A strain of *F. moniliforme* Sheldon was isolated from natural occurring field cases of ELEM along the Siskiyou Delta by Dr. B.J. Wilson, Vanderbilt University, Nashville, TN, submitted to the Northern Regional Research Center (NRRC) in 1967, and maintained as culture collection no. 6442. This organism was maintained on hay agar slants (6% hay, 2% DPG, 2% agar, pH 6-6.5) at the USDA NRRC, Peoria, Illinois. A steam-sterilized substrate, consisting of yellow corn (300 g) and 100 ml of distilled water in a Fernbach flask, was inoculated with 5 ml of a cell suspension of *F. moniliforme*. Sheldon prepared form a 7-day old PDA slant (potato-dextrose agar) and 5 ml of sterile distilled water. The inoculated substrate was incubated at 25°C as static cultures for four weeks, then dried in a forced air oven at 78°C for 3-4 hours. This cultured corn was ground in a Wiley mill using a 2 mm mesh screen.

Two apparently healthy 4-year old donkeys were housed in confinement within individual stalls. Both animals were gradually acclimated to a corn diet by the addition of whole yellow corn to the ration over a 8-day period. Alfalfa hay (1.0 kg) was provided daily throughout the study.

Donkey #1, 123 kg, was dosed through a stomach tube approximately every other day with the cultured corn at the rate of 7.6 g/kg body weight. Twelve doses, a total of 11.25 kg, of the cultured corn was given within a 27-day period. Tame feeding was discontinued after the 12th dose and the cultured corn was mixed in the ration of whole yellow corn (0.5 kg) and molasses at the rate of 4.8 g/kg body weight/day. Feeding was continued for 13 days with a consumption of 8.0 kg of cultured corn.

Donkey #2, 105 kg, was fed the cultured corn mixed in the ration of whole yellow corn (0.5 kg) and molasses daily for 16 days at the rate of 4.7 g/kg body weight. Total consumption of cultured corn was 24.8 kg.

Cultured corn was fed ad libitum to three New Zealand white rabbits for 29 days. The feed was provided in pellet form (50% cultured corn, 25% soybean meal, and 25% alfalfa meal).

Cultured corn (420 g) was extracted with 840 ml methanol-distilled water (40:60) for 3 minutes in a Waring blender. This procedure was done twice and the extracts were combined and dried to dryness in a forced air oven at 78°C. The residue was chromatographed on a chloroform/acetone/ethyl acetate mixture to recover moniliformin. The column was eluted with chloroform (500 ml), 5% methanol-chloroform (500 ml), and methanol (200 ml). Each eluate was concentrated to approximately 10 ml. Twenty percent of an eluate was added to 0.15 ml vegetable oil (Hunt-Wesson Foods, Inc., Fullerton, CA: Wesson Oil) and concentrated on a rotovap evaporator at 60° C. A 0.15 ml aliquot of the vegetable oil-elicuate mixture was intubated into the crop of six one-day old chicks. The chicks...
The animals in this study were necropsied immediately following death. All animals, except Donkey #1, were euthanized with pentobarbital sodium given intravenously. Tissue specimens were fixed in 10\% buffered formalin. Sections for microscopic examination were stained with hematoxylin and eosin.

RESULTS

Donkey #1 showed no clinical abnormalities during the first 38 days of feeding. On the 39th day he was unusually irritable and easily agitated. When removed from the stall for exercise he became refractory, hyperactive, incoordinated, and sweat profusely. He was put back into the stall and became extremely ataxic, fell into lateral recumbency, experienced extreme delirium and died five hours later. At necropsy the entire cerebral cortex was soft and friable. The left cerebral hemisphere contained a cavity, approximately 3.5 cm long, 2 cm wide, and 3 cm deep, filled with a yellowish-brown mucoid fluid. The cortex over the cavity was very thin and friable. Histopathologic examination of the cerebrum revealed congested vessels in the cortex and subcortical white matter. The subcortical white matter around the necrotic cavity was disrupted and rarefied. Neuraglia within these areas were pyknotic and eosinophilic. Perivascular hemorrhages were prominent in the subcortical white matter and gray matter. The liver had diffuse fatty vacuolated hepatocytes. (Figs. 1-3)

Fig. 1 Brain from donkey #1 with necrotic areas in both cerebral hemispheres. Cavitation in the left hemisphere (arrow) is evident.

Donkey #2 had signs of laminitis on the 14th day of feeding. All feed was withheld for 4 days and the condition improved. However, the animal developed a rear limb ataxia that remained throughout the study. The donkey experienced periodic anorexia of 1 to 2 day duration during the 46 days of

Fig. 2 Serial cross sections of brain from donkey #1 with prominent areas of necrosis and cavitation (arrows).

Fig. 3 Serial cross sections of the brain from a quarter horse that died in January 1979 from a naturally occurring outbreak of ELM in Illinois. Arrows indicate areas of necrosis.
No characteristic signs of ELEM were seen and the donkey was euthanized. No gross lesions of the brain or spinal cord were noted at necropsy. The mucosa of the duodenum was reddened and there were numerous petechial and ecchymotic hemorrhages on the surface and within the parenchyma of the testicle. Histopathologic lesions of the brain and spinal cord were unremarkable except for some mononuclear cell perivascular cuffing in the subcortical white matter of the cerebrum.

There were no signs of illness or any deaths in the rabbits or the day-old chicks. No significant gross or histopathologic lesions were seen in the rabbits, and postmortem examinations were not conducted on the chicks.

**DISCUSSION**

The results of this study are consistent with the clinical signs, gross and microscopic lesions of ELEM reported by previous authors. The ability to reproduce the syndrome in a donkey fed autoclaved corn cultured with *Fusarium moniliforme* has again established presumptive evidence that this fungus is involved in ELEM. However, the toxicant(s) responsible and the mode of action are not known. It is postulated that a toxic metabolite of the *Fusarium* spp. is responsible for the disease. Investigators in Japan isolated 26 *Fusaria* spp. from bean-hulls used as feed and bedding in cases of equine deaths that were characteristic of ELEM (14). Sixteen of the isolates were lethal to mice in toxicity tests and 90% of these isolates produced trichothecone-type mycotoxins. *F. moniliforme* was one of the species found, but no trichothecones were isolated from it. *Moniliforme* is often reported to be the toxic principle of ELEM. A previous study used chicks as a bioassay model for toxicity testing of moniliforme (5). The extracts from the cultured corn used in this study were not lethal to chicks in doses comparable to those which produced death in the study cited. Hence, the chick bioassay results indicate the absence of a monovalent salt of moniliforme in the cultured corn.

**SUMMARY**

Signs and lesions characteristic of equine leukoencephalomalacia were produced in one of two donkeys given corn cultured with *Fusarium moniliforme* Sheldon. Gross and histopathologic lesions of the cerebrum included an extensive necrotic cavitition within one cerebral hemisphere, disruption and rarefication of the subcortical white matter, prominent perivascular hemorrhage, and some mononuclear cell perivascular cuffing. Another donkey and three rabbits fed the cultured corn did not develop characteristic signs or lesions of the toxicosis. Chick bioassay studies indicated that the cultured corn which produced the disease did not contain a monovalent salt of moniliforme.

**REFERENCES**


6. Graham, R: Results of Inoculating Laboratory Animals with Equine Brain-tissue Suspensions and Equine Brain-tissue Filtrates from Sterile Cases of So-called Cornstalk Disease. JAVMA 86:778-780, 1935.


