

## New Information on the Lifecycle of *Sarcocystis neurona*

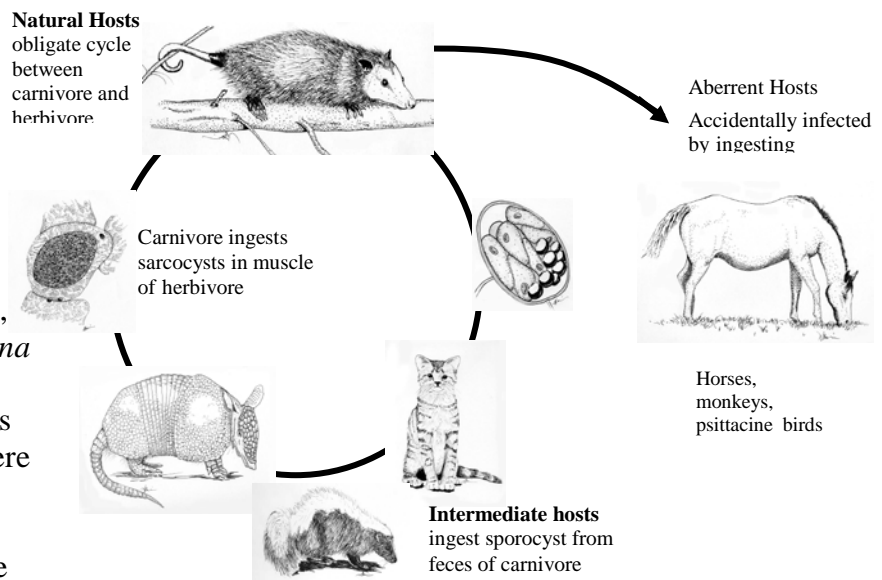
The parasite *Sarcocystis neurona* is the primary cause of equine protozoal myeloencephalitis (EPM) in horses, which is a serious, often life-threatening neurological disease. This article deals with new information on the lifecycle of the parasite.

All members of the group *Sarcocystis* parasites require at least two separate hosts for their development (Figure 1). These parasites reproduce (sexually) in the intestinal wall of one animal (a carnivore), ultimately producing sporocysts that are passed in the feces. Ingestion of these sporocysts in the environment by another animal (a prey animal) continues the life cycle. Once *Sarcocystis* sporocysts are eaten by the prey animal, the parasite breaks out and gets into the blood stream and then into the muscles or other body tissues where they reproduce (asexually) to high numbers. They ultimately form muscle cysts that are infective to the carnivore host when they are eaten. Thus, the parasite cycles between the carnivore and the prey animal.

Opossums are the natural carnivore host for *Sarcocystis neurona*. At this time, they are the only known carnivore, which sheds *Sarcocystis neurona* sporocysts in the feces. The prevalence of *Sarcocystis neurona* in opossums in Michigan and the Great Lakes area appears to be high. Horses are accidentally infected with this parasite when they graze pastures contaminated by infected opossums. In Michigan, the prevalence of antibodies to *Sarcocystis neurona* in horses indicating exposure to the parasite was determined to be 60% (Rossano et al., 2000). It is the accidental hosts, like the horse, that have severe disease due to the parasite. From our work, exposure to opossums is the major risk factor for horses for development of EPM. More horses are infected in areas with many opossums.

Three wild animal species, the nine-banded armadillo, the striped skunk, and the raccoon have recently been identified as prey hosts of *Sarcocystis neurona*. These animals are infected by ingesting *Sarcocystis neurona* sporocysts in opossum feces. Once they eat the sporocysts, they go on to develop cysts in their muscles. To continue the cycle, the muscles with cysts are eaten by opossums and the parasite once again develops in the intestines of the opossum. The prevalence of *Sarcocystis neurona* infection in opossums in a given area is dependent upon the availability of infected carrion or prey, and the armadillo, the skunk, and the raccoon may be significant sources of *Sarcocystis neurona* in the form of road-killed carcasses. To be a source of infection to opossums, armadillos, skunks and raccoons must have the opportunity to be infected with sporocysts from opossum feces and subsequently be available to be consumed by opossums. It is clear that armadillos are an important prey species host for *Sarcocystis neurona* in the South, while raccoons and skunks are likely to be more important in the North.

Figure 1: Life cycle of *Sarcocystis neurona*



A recent study by scientists at USDA and The Ohio State University showed that domestic cats could serve as prey hosts to *Sarcocystis neurona* when they eat *Sarcocystis neurona* sporocysts from infected opossums. Muscle cysts produced in these cats were infective to opossums and were subsequently confirmed to be *Sarcocystis neurona*. This phenomenon has been shown in a feral cat as well. This cat had muscle cysts that were positively identified as *Sarcocystis neurona* using several tests (Turay, et al.).

Studies conducted by the Michigan State University EPM Research Group addressed the role that domestic cats play in the natural life cycle of the *Sarcocystis neurona*. It seemed plausible to us that in a state with relatively high *Sarcocystis neurona* exposure of horses (like Michigan) that there may also be a significant number of exposed cats, if cats act as common prey host. The purpose of our study was to estimate the exposure by detecting antibodies in the blood stream of cats in Michigan by testing banked serum samples from cats stored at the Michigan State University, Animal Health Diagnostic Laboratory. 10/196 (5%) of the cats tested positive for antibodies to *Sarcocystis neurona*. We conclude that domestic cats are not likely to play a substantial role as prey hosts in the natural life cycle of *Sarcocystis neurona*. Our study does demonstrate that natural infection of domestic cats may occur, and small animal practitioners should be aware of this when cats with neurological disease are presented to their clinics.

Horse owners should recognize that exposure to opossums poses the major threat to their horses. Exposure to armadillos, skunks, raccoons and especially cats is unlikely to pose a problem. Horse owners with previous problems due to EPM may consider an opossum control program on their farms. Submitted by Linda Mansfield, VMD, PhD and Mary Rossano, M.S., Population Medicine Center, Department of Large Animal Clinical Sciences, College of Veterinary Medicine, Michigan State University, East Lansing, MI. □

## Prevention of EPM

### Strongid C Kills *Sarcocystis neurona* Merozoites

A safe prevention strategy is needed to protect horses from infection with the parasite that causes EPM. EPM is considered to be the most important neurological disease of horses in North, Central, and South America. Exposure to the

causative agent of EPM, *Sarcocystis neurona*, is high with infection in horses in some areas exceeding 50%. This high exposure rate, in addition to the seriousness of the disease, supports the need for prevention, rather than treatment, as a method of controlling the disease. Daily feeding of an effective preventative drug would be a possible means of protecting horses from infection with *Sarcocystis neurona* and subsequent development of EPM.

It has been speculated that pyrantel tartrate, the active ingredient of Strongid® C (Pfizer, Inc., New York City, NY), may have activity against *Sarcocystis neurona*. Strongid® C is a daily anthelmintic feed supplement used to protect horses from strongyle worms. Theoretically, daily pyrantel tartrate administration could prevent *Sarcocystis neurona* infection in equids by killing sporozoites as they hatch out in the gut. Studies done by Elizabeth Kruttlin, DVM student at MSU in the laboratory of Linda Mansfield, have tested the efficacy of the active ingredient of Strongid C against one stage of the parasite, the merozoite. These tests showed that the drug kills this stage of the parasite at the doses comparable to that given to horses. However, this is the stage in the brain and spinal cord of horses, not the stage found in the intestinal tract. Also, these studies were done in tissue culture in the laboratory and, as such, are preliminary in nature. Strongid C will need further testing as a preventative drug by feeding it to horses living in an area with *Sarcocystis neurona*.

To understand how preventative drugs must be given and must act to prevent *Sarcocystis neurona* infection, we need to understand how the parasite is acquired by the horse. *Sarcocystis neurona* is acquired by horses by eating sporocysts from grass or soil contaminated with opossum feces. It is likely that sporozoites hatch out of sporocysts in the digestive tract of the horse, penetrate the gut wall, and travel to the central nervous system of some horses where they are found as merozoites. The merozoite stage of the parasite replicates in the neurons of the brain and spinal cord leading to the clinical signs of EPM. So we need a drug that can kill the sporozoites after they hatch out and before they penetrate into the tissues of the gastrointestinal tract and spread.

The purpose of this study was to determine if pyrantel tartrate has activity against *Sarcocystis neurona* merozoites. The merozoite stage of the

parasite used in this study replicates in the CNS of horses and is not likely to be exposed to pyrantel tartrate, a drug that has low absorption and remains primarily within the digestive tract. However, it is believed that results attained from merozoite studies may be useful to model how the drug will work against the sporozoite stage of *Sarcocystis neurona* found in the digestive tract. Positive results would lead to consideration of further studies investigating the possibility of using Strongid® C as a preventative for EPM in horses.

In our study, *Sarcocystis neurona* merozoites were exposed to a range of concentrations of pyrantel tartrate or sodium tartrate (a control) between 0.001M and 0.01M (Kruttlin et al., 2001). Merozoites were then placed onto equine dermal cell cultures and incubated for two weeks to check for viability. At one and two weeks post inoculation, plaques produced by the growing parasites were counted and the counts compared between treatment groups and controls using the appropriate statistical methods. Merozoites exposed to concentrations of pyrantel tartrate higher than 0.0025M (8.91X10<sup>-4</sup> g/ml) did not produce plaques in equine dermal cells while those exposed to similar concentrations of the tartrate salt or medium alone produced significant numbers of plaques. These results demonstrate that Strongid C (pyrantel tartrate) appears to be 100% lethal to *Sarcocystis neurona* merozoites at these concentrations. This result has prompted further investigations into the possibility of using Strongid® C as a preventative for EPM in horses.

Horse owners should recognize that there were limitations to this initial study. The merozoite stage of *Sarcocystis neurona* is found in the brain and spinal cord of horses and is not exposed to pyrantel tartrate, a drug that remains primarily in the digestive tract. However, because the merozoite stage is easy to get and has some similarities to the sporozoite stage found in the gut of the horse, we elected to initially study the effects of pyrantel tartrate on this stage of the parasite. Further studies are underway to test the drug in horses against the sporozoite stage in a natural farm setting with no injury to the test subjects. We will let the horse community know right away, if this drug is effective as a preventative in a real life situation. Submitted by Linda S. Mansfield, M.S., V.M.D., Ph.D., Departments of Large Animal Clinical Sciences and Microbiology, College of Veterinary

Medicine, Michigan State University, East Lansing, MI. Based on a study recently published Kruttlin E, Vrable R, Murphy AJ, Rossano MG, Mehler S, and Mansfield LS. 2001. The effects of pyrantel tartrate on *Sarcocystis neurona* merozoite viability. *Veterinary Therapeutics* 2(3):268-276. □

## Treatments for EPM: What's New for 2002?

### Marquis® - The first drug approved for treatment of EPM

In July, 2001 Bayer released Marquis®, the first drug to be licensed for treatment of horses afflicted with neurologic disease attributable to EPM. Marquis® is formulated as a paste (like a dewormer) with 7 doses, 5 mg/kg dose administered in the mouth once daily, in each paste syringe. The recommended treatment duration is 28 days (4 paste syringes) and costs between \$600-800, depending on the clinic through which it purchased (it is only available through your veterinarian).

The active drug in Marquis® is ponazuril, a metabolite (totrazuril sulfone) of totrazuril (Baycox®) that has been demonstrated to kill *Sarcocystis neurona* merozoites (the form of the parasite infecting neural tissue in horses) in the test tube. Studies by Bayer have also shown that ponazuril is absorbed across the intestinal tract and reaches effective concentrations in both blood and spinal fluid (site where drug is needed). Further, the drug appears to be relatively safe for horses although an occasional horse may experience GI upset during the first few days of treatment. GI upset may be manifested as colic or diarrhea due to simultaneous killing of important protozoa in the horse's large intestine. Because of the risk of GI upsets, some veterinarians recommend prophylactic use of Banamine® during the first 5-7 days of treatment with Marquis®. Finally, when horses were administered 6 times the recommended dose (30 mg/kg) for twice the recommended treatment duration, few adverse effects were observed although a few animals had transient diarrhea and some swelling of the inner layers of the uterus were detected at autopsy examination. Potential adverse effects on reproduction have not been studied and Marquis® has not been licensed for use in pregnant or lactating mares.

In order for Marquis® to be licensed, Bayer sponsored a field efficacy study in which 101 horses diagnosed with EPM were treated for 28 days and

follow-up examinations and spinal taps were performed 3 months after treatment ended. A successful response to treatment was defined as either a one grade (minimum) improvement in the neurologic deficits (graded on a scale of 0-5) or a negative spinal fluid western blot test result found 90 days after cessation of treatment. Overall, 62% of treated horses showed a positive clinical response and 10% had developed a negative spinal tap western blot test result 3 months after treatment ended. It is important to recognize that clinical improvement did equal a complete return to normal.

So, does Marquis® work better than other drug combinations for treatment of EPM? That remains an unanswered question. The results of the Marquis® field study (that about 60% of horses respond to treatment) are similar to what has often been described for the more traditional combination treatment of sulfadiazine/pyrimethamine for 90-180 days. It is important to remember that true EPM is a bad disease and that only about 50% of affected horses can be expected to show a full response to treatment. Anecdotally, some veterinarians have observed more rapid improvement with Marquis® than with sulfadiazine/pyrimethamine combinations. Unfortunately, none of the drug studies performed to date have compared the response to treatment with different drugs – all we have are lots of opinions based on anecdotes and individual case experiences.

Finally, a disconcerting observation in several experimental infection studies with *Sarcocystis neurona* has been that when horses do develop neurologic disease (an inconsistent occurrence) after being dosed with *Sarcocystis neurona* sporocysts by a stomach tube, the course of the neurologic disease has often been self-limiting with improvement observed without any treatment. These observations call into question the results of all drug studies performed to date because none of the studies has incorporated a comparison group of non-treated or placebo-treated horses.

Thus, although we now have a drug licensed for treatment of horses with neurologic disease attributable to EPM, we do not really know if this drug is any better than previously used treatments. However, the Marquis® studies did provide additional useful information. Specifically, it was found that several days of treatment were needed to achieve adequate blood and spinal fluid concentrations of the drug. Thus, intermittent use

(e.g., weekly or biweekly) of Marquis® or the similar drug Baycox® as a potential preventive (and less expensive) treatment makes little sense and is likely a waste of money because the drug concentrations reached in blood and spinal fluid are inadequate. *Submitted by Harold Schott, D.V.M., Ph.D., Diplomate ACVIM, Department of Large Animal Clinical Sciences, College of Veterinary Medicine, Michigan State University, East Lansing, MI.* □

## Vaccination for EPM in Horses

### The EPM Vaccine: What do we know about it?

In late 2000, Fort Dodge Animal Health was granted a conditional license for their vaccine against *Sarcocystis neurona*, the most common protozoal organism causing EPM. Conditional licensure is granted by the USDA in situations for which a disease is of high concern to the horse industry (true for EPM) and for which the vaccine has a “reasonable expectation of efficacy”. What that means is that the vaccine has been shown to have an effect (basically that it produces serum antibodies against *Sarcocystis neurona* when given to horses) but there is no evidence at present that development of these antibodies will prevent infection in horses. Further, before a conditional license is issued vaccine safety has to be well documented. Fort Dodge Animal Health is actually to be congratulated for their efforts to document safety of the EPM vaccine because they administered two doses to almost 1000 horses (most safety studies are performed on 200 or fewer horses). Importantly, none of the vaccinated horses developed neurological disease attributable to EPM and the rate of adverse effects (local swelling, lethargy, fever, etc.) was low (1-2%).

You may be asking: “How can a vaccine company can get away with selling a product like that?” Well, it’s not the first time and it certainly won’t be the last. Fort Dodge Animal Health is simply responding to the overwhelming demand of the horse industry for measures to prevent this devastating neurological disease of horses. One of their previous products – the rotavirus vaccine given to pregnant mares – was also initially released with a conditional license and it still has a conditional license several years later because attempts to prove vaccine efficacy under field conditions have failed. Until we demand greater efficacy of the products we are willing to give to

our equine companions, the situation is not likely to change very quickly – just think of how many people are willing to give the much more poorly regulated nutraceuticals to their horses (or even take them themselves).

Fort Dodge Animal Health is also actively trying to figure out whether the vaccine is effective. But there is a major obstacle – lack of an experimental model of EPM in horses. Because researchers can't consistently reproduce EPM by giving *Sarcocystis neurona* sporocysts (the form of the parasite shed in opossum feces) to horses, it is currently impossible to determine the efficacy of the vaccine to prevent EPM (the same problem holds true for drugs used to treat the disease). However, hope is on the horizon because researchers appear to be closing in on developing such a model. In the meantime, a 3-year epidemiologic, field efficacy study was initiated in September, 2001 to try to answer the question. This study is using horses from 12 equine hospitals around the United States, including MSU. It also warrants mention that not all vaccines have been demonstrated to be effective in a disease challenge model. The best example is vaccines against Rabies – no human challenge studies have ever been (or will likely ever be) performed because infection with the Rabies virus is highly fatal.

If you are considering having the vaccine administered to your horse, you should also read the product label. Specifically, it is labeled “For vaccination of healthy horses as an aid in the prevention of neurologic disease (EPM) caused by subsequent exposure to the protozoan *Sarcocystis neurona*.” The “as an aid” clause has to be included because efficacy data is lacking (governmental regulation). More importantly, this statement actually says that vaccine use would likely be more effective in horses that have not been exposed to *Sarcocystis neurona* (i.e., in horses without detectable serum antibodies = negative serum western blot test results). Thus, the product is not really targeted for horses that already test positive for exposure to this parasite (50-60% of all horses in the mid-West) but would be more appropriate to use in a horse being moved from an opossum (and EPM) scarce area (e.g., Montana) to an opossum (and EPM) haven like Michigan. Unfortunately, this aspect of vaccine use has been rather poorly presented to veterinarians and horse owners. Further, the vaccine has no use as a treatment for

horses already afflicted with neurologic disease attributable to EPM. A final concern about vaccine use is that it could interfere with using the current western blot as a diagnostic tool. Ongoing collaborative work between MSU and Fort Dodge Animal Health is addressing this issue.

So where does that leave us? Basically, the EPM vaccine is a product that is safe to give to horses but we do not know if it is effective. Veterinarians should present their clients with this information in order for horse owners to make a knowledgeable decision about whether or not to have the vaccine administered to their horse. At MSU, we remain neutral on the vaccine – we neither recommend nor discourage its use.  
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## Differentiating EPM from West Nile Virus: A Job for Your Veterinarian

### Differentiating EPM from West Nile Virus

Diagnosis of EPM in horses was never an easy task for the veterinarian. With the introduction of West Nile virus into the US, veterinarians will be faced with an even more complicated problem in diagnosing neurological disease in horses. More importantly, West Nile virus can infect humans, while *Sarcocystis neurona* can only infect animals. It will be important for veterinarians, public health officials, and horse owners, among others, to keep abreast of new information about both of these pathogens.

### West Nile Virus

Ohio State authorities say “West Nile virus is a mosquito-borne virus that can cause encephalitis (inflammation of the brain) or meningitis (inflammation of the lining of the brain and spinal cord). West Nile virus is transmitted by the bite of an infected mosquito. It is widespread in Africa, southern Europe, and western Asia. It first appeared in the United States in 1999 in the greater New York City area. In 2000, it spread to all of the New England states and south to North Carolina. It has caused illness and mortality in humans, wildlife and domestic animals, especially birds and horses. In humans, it causes an influenza-like illness that may lead to aseptic meningitis, encephalitis, and death, especially in persons over 50 years of age. West Nile virus is important because it affects not

only people, but also wildlife (including many game animals) and some domestic animals. Clinical signs of West Nile Virus infection in horses include: listlessness, stumbling, lack of coordination, ataxia, partial paralysis, and death. Horses with West Nile Virus often do not have a fever.”

Horses are known as a dead-end host of West Nile Virus, that is, they can become ill with West Nile Virus, but they do not maintain sufficient virus in the blood to infect either other mammals (including humans) or mosquitoes. Surveillance of horses has occurred in areas currently affected with West Nile Virus and in additional States on the eastern coast of the United States to monitor the possible spread of WNV. This surveillance consists of investigating suspect cases in horses. Because horses are not known to play a role in transmission of WNV, quarantines were never placed on any non-clinically ill horses in the outbreak area. However, some foreign countries have banned the importation of horses from New York.

The Michigan Department of Community Health and Michigan Department of Agriculture have information on West Nile virus that is specific for the State of Michigan that can be found at the following websites (#1 MDA: <http://www.mda.state.mi.us/consumer/westnilevirus/> , and #2 MDCH: [http://www.mdch.state.mi.us/sub/community\\_public\\_health/index.htm](http://www.mdch.state.mi.us/sub/community_public_health/index.htm), and

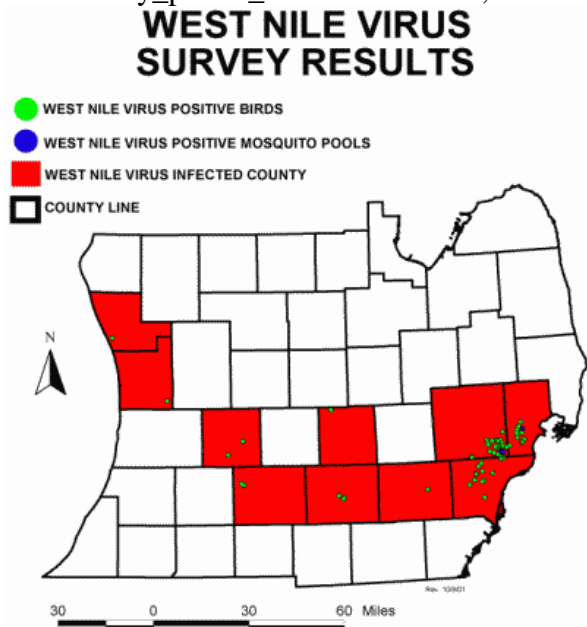


Figure 2: Reprinted with permission from the Michigan Department of Agriculture website.

#3 [www.michigan.gov/mda](http://www.michigan.gov/mda), then follow the links consumer information, animal health and West Nile virus.

### Equine protozoal myeloencephalitis (EPM)

Equine protozoal myeloencephalitis (EPM) is a neurological disease of horses caused by *Sarcocystis neurona*. *Sarcocystis neurona* is a protozoan parasite that replicates in cells of the central nervous system (Mackay, 1997). When the parasite gains access to a horse's spinal cord, gait abnormalities develop as one or more limbs become weak (paresis) and movement becomes incoordinated (ataxia). If the parasite enters the brain, affected horses may develop a head tilt, lip or ear droop or they may have difficulty swallowing. More serious signs can include severe weakness leading to recumbency, seizures and even death. Subtle gait deficits in athletic horses are not only frustrating to their owners; they are equally frustrating diagnostic challenges to equine veterinarians. When a definite diagnosis remains elusive, equine protozoal myeloencephalitis (EPM) usually gets added to the list of possible causes. Unfortunately, a definitive antemortem diagnosis of EPM is difficult.

For further information about these diseases you may contact your state veterinarians office. An excellent fact sheet is available at the following website: <http://www.odh.state.oh.us/odhprograms/zoodis/wnv/Pubs/wnvhorseown.pdf> that was prepared by the following groups: Ohio Department of Health, the Ohio Department of Agriculture, the Ohio Department of Natural Resources, The Ohio State University, the Ohio Environmental Protection Agency, the Association of Ohio Health Commissioners, the Ohio Mosquito Control Association, the Ohio Environmental Health Association, and the United State Department of Agriculture. Submitted by Linda S. Mansfield, M.S., V.M.D., Ph.D., Departments of Large Animal Clinical Sciences and Microbiology, College of Veterinary Medicine, Michigan State University, East Lansing, MI from materials provided by MDCH, MDA, ODH, and ODA. □

## EPM Newsletter

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