Equine Viral Encephalomyelitis in Canada: A Review of Known and Potential Causes

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Abstract

Rabies, equine herpesvirus type I, and eastern and western encephalomyelitis viruses, known causes of equine neurological disease, are reviewed with emphasis on epidemiology, pathogenesis, clinical signs, and diagnosis.

Several arboviruses known to be active in Canada and capable of producing neurological disease in humans (Powassan, St. Louis encephalitis, snowshoe hare, and Jamestown Canyon viruses) are discussed as potential causes of encephalomyelitis in horses.

Key words: Equine, encephalomyelitis, viral, arbovirus.

Can Vet J 1987; 28: 497-504

Résumé

Une revue des causes connues et potentielles d'encéphalo-myélite équine virale, au Canada

Les auteurs présentent une revue de la littérature relative au virus de la rage, à l'herpèsvirus équin #I, ainsi qu'aux virus de l'encéphalo-myélite équine de l'est et de l'ouest, qui représentent autant de causes connues de maladies neurologiques équines, en insistant sur l'épizootiologie, la pathogénèse, les signes cliniques et le diagnostic.

Ils commentent aussi, à titre d'agents étiologiques potentiels d'encéphalo-myélite chez les chevaux, plusieurs arbovirus reconnus comme actifs au Canada et capables de produire une maladie neurologique chez les humains, entre autres le virus Powassan, ainsi que ceux de l'encéphalite de St-Louis, du lièvre et du canyon de Jamestown.

Mots clés: équin, encéphalomyélite, viral, arbovirus.

Introduction

Many diseases are known to produce central nervous system (CNS) disturbances in the horse, the differential diagnosis of which have been described elsewhere (1). A fatal nervous disease of horses was reported along the eastern coast of the United States as early as 1831 (2). Since that time several viral agents have been discovered which are capable of causing encephalomyelitis in the horse (3). Unfortunately the isolation of a viral agent from the brain of an affected animal is often unsuccessful. Rabies is the only virus that is readily and reliably demonstrated in brain tissue of affected horses, although equine herpesvirus type I (EHVI), eastern equine encephalomyelitis (EEE) and western equine encephalomyelitis (WEE) viruses have been isolated from brain tissue of clinically affected animals (4, 5, 6, 7).

More often however, a presumptive diagnosis of viral encephalomyelitis is made on the basis of the clinical history, serology, and histological lesions of a nonsuppurative encephalomylitis, one having eliminated other causes of CNS disease.

Eighteen arboviruses have been isolated in Canada (8), several of which have the potential to cause disease in humans (9). These include EEE, WEE, Powassan (POW), St. Louis encephalitis (SLE), snowshoe hare (SSH), Jamestown Canyon (JC), and Colorado tick fever viruses.

The purpose of this paper is to review the role of rabies, EHVI, EEE, and WEE viruses in equine neurological disease. Powassan, SLE, SSH, and JC viruses are also discussed as possible causes of encephalomyelitis in horses.

Rabies

Rabies is caused by a single-stranded RNA virus of the Rhabdoviridae family (10). The disease is transmitted to horses by inoculation of the virus into a wound; such a wound is usually inflicted by the bite of a rabid animal, e.g. fox, dog, cat. The incidence of equine rabies in Canada is low (Table I - Agriculture Canada data).

Rabies is a neurotropic virus which replicates first in myocytes near the site of inoculation and then enters local neuromuscular and neurotendinous spindles (11). The virus travels up peripheral nerves, probably in the axoplasm, to the dorsal root spinal ganglia and from there invades the CNS (11). After invasion of the CNS, virus is rapidly disseminated, with early selective infection of neuronal populations (12). Rabies virus is then transmitted centrifugally along all peripheral nerves (13), and in this way leads to salivary gland infection. Virus replicates in acinar cells with budding of virions directly into the salivary ducts (14).

The incubation period of rabies in the horse is variable, ranging from two weeks to three months (15, 16). It depends on the dosage and strain of infecting virus, site of inoculation, and perhaps, the period of dormancy in myocytes at this site (11, 17, 18). In contrast to the frequently long incubation period, the clinical course of the disease is short, with most animals dying within ten days of the onset of illness (3).

TABLE I
Number of Equine Rabies Cases Diagnosed in Six Canadian Provinces from 1978-1985

Province	1978	1979	1980	1981	1982	1983	1984	1985
Quebec	3	1	2		1			
Ontario	24	23	33	24	31	13	7	25
Manitoba	4		1		1	1	1	
Saskatchewan	1				2	1	1	3
Alberta					1			
British Columbia							1	

Data from Agriculture Canada — Food Production and Inspection Branch

The clinical signs of equine rabies are extremely variable and include: hyperesthesia, recumbency, fever, convulsions, behavioral change, ataxia, incontinence, dysuria, muscle fasciculations, depression, anorexia, lameness, paresis, sweating, hyperexcitability, colic, and cranial nerve deficits (15, 16, 19). The histological lesions when present, are typical of a nonsuppurative meningoencephalomyelitis and ganglioneuritis; i.e. meningitis, lymphocytic perivascular cuffing, glial nodules, and ganglion infiltration with satellitosis and neuronophagia (20, 21). Negri bodies are specific for rabies and are best demonstrated in Purkinje cells of the cerebellum, neurons in the hippocampus, and the pyramidal cells of the cerebral cortex. They are found less frequently in the neurons of the thalamus, pons, medulla, spinal cord and sensory ganglia (22). However, in extensive surveys, Negri bodies were not found in 10-15% of rabies cases which were proven positive by the mouse inoculation test (4, 18, 23).

The fluorescent antibody (FA) test is the most commonly used method of detecting rabies virus, as it is rapid and identifies up to 99.8% of infected brain specimens (24, 25). Fluorescent antibody tests have also been used to detect rabies virus antigen in nerves in facial skin biopsies of mice (26, 27), skunks (28), and a calf (29) prior to the development of clinical signs. This method of antemortem diagnosis has not been reported in horses, nor is it likely to become an important diagnostic tool for equine rabies.

The mouse inoculation test is often used in combination with the FA test (30). Fluorescent-positive, mouse-negative specimens can occasionally be expected, since the FA test detects inactivated as well as live antigen. In this respect, fluorescence tests are more sensitive than tests in mice (25). Rarely, mouse-positive, fluorescent-negative specimens occur; this may result from an error in setting up or reading the FA test or of insufficient sampling of brain tissue (25).

Virus neutralization of brain tissue is a valuable but uncommon method of diagnosing rabies in those animals with nonsuppurative encephalitis for which fluorscent antibody and mouse inoculation tests are negative (31). In such cases high levels of rabies antibody in the brain may have masked the viral antigen present.

Monoclonal antibodies have been used to distinguish strains of rabies virus (32, 33, 34), however, the expense involved means that this method of testing will not likely be used as a routine diagnostic tool in the near future.

Recently a new technique for the diagnosis of rabies has been reported (35). This method involves inoculating cell cultures with brain or spinal cord tissue suspected to contain rabies virus, and examining the cell cultures four or five days later by direct immunofluorescence. This test is relatively rapid and very sensitive. In conjunction with FA examination of brain sections, it is now being used routinely in the diagnosis of rabies in Canada.

In Canada there are currently two vaccines available for intramuscular use in horses. Both of these are killed virus vaccines grown in hamster kidney cells (Rablan, Langford Laboratories Ltd., Guelph, Ontario and Imrab, MTC Pharmaceuticals, Mississauga, Ontario). It is recommended that these be administered to horses annually in areas in which rabies is prevalent.

Equine Herpesvirus Type I

Although the two main syndromes of EHVI infection are upper respiratory tract disease and abortion, there have been many reports worldwide of neurological disease associated with EHVI (36, 37, 38, 39, 40). Clinically affected animals may have an acute onset of paresis, ataxia and/or paralysis during or after epizootics of abortion or upper respiratory tract infection of in-contact animals.

Bitsch and Dam (41) define three grades according the degree of paralysis:

Grade 1:
Slight ataxia which may persist for two to seven days after which recovery is complete.

Grade 2:
Inability to rise unassisted, severe incoordination of the hind limbs, and unwillingness to move. Urinary incontinence is frequently seen. Horses may regain full coordination after a protracted recovery.

Grade 3:
Inability to stand unassisted, but continue to eat and drink normally. The prognosis is very poor in cases of extensive paralysis, and death may occur. Incoordination of the hind limbs may still be seen after several months in those animals which do recover.

In addition, some animals may develop profound central nervous signs with convulsions and death.

Histological lesions in affected horses are confined to the brain, spinal cord, and trigeminal ganglia (36, 37, 40, 42, 43). Vasculitis with secondary hypoxic degeneration in adjacent neural tissue is the prominent histological change in both natural and experimental cases. Vascular changes are more prominent in the spinal cord than in the brain, and white matter lesions of the spinal cord are more prominent than gray matter lesions. There is necrotizing fibrinoid vasculitis with degeneration of the media and/or adventitia, endothelial swelling and proliferation, hyaline thrombosis, and perivascular mononuclear cuffing. Small arteries, arterioles, and venules are predominantly affected. Hemorrhage is sometimes seen and is probably secondary to the vascular lesions. Gasserian ganglioneuritis is often present and is characterized by infiltration of large numbers of lymphoid cells between neurons, proliferation of capsule cells, and occasionally hemorrhage. Neither inclusion bodies nor neuronophagia are present, which is in striking con-

trast to the pathology seen with other forms of encephalitis associated with the herpesviruses. Herpes simplex encephalitis in man (44), infectious bovine rhinotracheitis in calves (45), Aujeszky's disease in piglets (46), and canine herpesvirus in puppies (47, 48) all have clear evidence of primary neurotropism with virus multiplication in neurons (inclusion bodies) and neuronophagia. This distinct difference between herpesvirus infection in the CNS of horses and herpesvirus infection in the CNS of other species suggests that a different pathogenesis is involved.

Diagnosis in the living animal cannot be made definitively although a history of respiratory tract disease, particularly in young stock, and/or abortion in the last trimester of in-contact animals, should suggest herpesvirus encephalomyelitis as a possible cause of nervous disease in those animals with paresis or paralysis (38). This historical evidence is further supported by serological evidence of infection with a fourfold or greater rise in serum antibody titer to EHVI (42). There may be no gross postmortem findings but close inspection may reveal rare radicular and focal spinal cord hemorrhages. This together with the characteristic histological lesions of a necrotizing vasculitis and focal malacia as described earlier further support the diagnosis.

It is possible that the lesions of the CNS are due to a direct action of EHVI since this virus has been cultivated from the nervous tissue of horses exhibiting neurological disease on several occasions (5, 36, 38, 39, 49). However, in most cases, examination of brain and spinal cord has failed to demonstrate EHVI antigen (40, 41, 42, 50).

A number of hypotheses have been proposed to explain the failure to demonstrate the virus: specialized methods of viral isolation may be required (40); strain peculiarities may exist (40); and virus neutralizing antibody may mask the virus and viral antigen (37, 40, 42).

The viremia of EHVI is strictly cell-associated (51), the virus being capable of spreading directly from an infected cell to contiguous cells without an extracellular phase. This may explain the occurrence of the disease in animals with preexisting virus neutralizing anti-EHVI antibody titers, and the primary vascular nature of the lesion, the virus infecting endothelial cells directly from the blood leukocytes which serve to transport the pathogen. This hypothesis complements the idea of an immune-mediated component to the disease with infection being superimposed on a sensitized system resulting in an anamnestic response. The antibodies which are produced in large quantities may interact with the antigen in endothelial cells causing a necrotizing vasculitis. Further parenchymal damage occurs as a result of ischemia (36, 40). Nevertheless, viewed in relation to the ubiquitous occurrence of EHVI infection in horse populations, outbreaks of paralysis are extremely rare and the specific circumstances and factors which predispose to their occurrence are at present undefined.

Two vaccines are currently available in North America to immunize horses against EHVI — an intramuscular, modified live vaccine produced on an equine cell line (Rhinomune, Norden Laboratories, Lincoln, Nebraska) and a hamster adapted preparation of killed EHVI (Pneumabort, Fort Dodge Laboratories, Fort Dodge, Iowa). Neither of these vaccines can be used to protect specifically against the neurological form of the disease because, as yet, a neurotropic strain of EHVI has not been recognized.

Another vaccine containing a modified live virus adapted to a monkey cell line (Rhinoquin, Bio-Ceutic Laboratories, St. Joseph, Maryland) was withdrawn from the commercial market in 1978 as the result of its association with posterior paralysis in horses (52, 53). There have been no reports of neurological disease associated with the use of the modified live vaccine produced in an equine cell line.

Eastern Equine Encephalomyelitis

Eastern equine encephalomyelitis was first recognized as a distinct disease in the United States in 1933 (6, 54). It has since caused intermittent outbreaks among horses, humans, and pheasants in various sections of the eastern USA, mainly in the coastal states from Massachusetts to Texas (55). Eastern equine encephalomyelitis is caused by a single-strand, enveloped, RNA virus of the genus Alphavirus, family Togaviridae (56, 57).

Eastern equine encephalomyelitis virus is believed to be maintained in a primary enzootic cycle involving small birds and *Culiseta melanura*, a swamp mosquito that does not feed on large birds or mammals (55). With alterations in the conditions of the marshes or swamps, changes in rainfall, different susceptible bird populations, and changes in mosquito breeding, the virus can spill over into other mosquito hosts, e.g. *Aedes* spp, that do feed on mammals. Only then are cases of encephalomyelitis seen in horses.

No epidemics of EEE have yet been recorded in Canada, but a small outbreak occurred in horses in 1938 in the vicinity of St. George, Ontario (58) and an equine epizootic occurred in the Eastern Townships of Quebec in 1972 (59, 60, 61).

Horses exposed to an infectious dose of EEE respond in one of the following ways (62): 1) animals may experience an inapparent infection; 2) infected animals may develop high fever and viremia which passes without subsequent CNS disease; or 3) animals may develop clinical signs of neurological disease. Affected animals may exhibit depression, incoordination, circling, ataxia, head pressing, continuous chewing movements, hyperexcitability, convulsions, and/or prostration. Results may be fatal or nonfatal, with or without sequelae.

Histological lesions fall into two general categories (63, 64). In horses dying within a day of the onset of nervous symptoms, there is a diffuse infiltration of the gray matter with polymorphonuclear leukocytes (PMNs). Vascular changes are prominent, with endothelial swelling, thrombus formation, and persivascular infiltration of lymphocytes and PMNs. Neuronal damage is mild. In animals surviving two days or longer, PMNs are no longer present. They are replaced by diffuse microgliosis and nodules consisting of large mononuclear cells and microglia. Perivascular cuffs at this stage are almost exclusively lymphocytic. Inclusion bodies are not present.

The diagnosis of EEE in horses is made by a combination of clinical findings, epidemiological information (cases occur during the late summer when mosquito populations are high), histopathology, serology, and viral isolation.

Hemagglutination-inhibition (HI) and virus neutralizing (VN) antibodies appear in the serum four to six

days after infection (62, 64) and a fourfold increase in either of these serum antibody titers in paired sera may be considered presumptive evidence of infection. The complement-fixation (CF) test has not proved to be reliable in demonstrating recent infections (64).

Viral isolation provides a definitive diagnosis of EEE and is most likely to be successful if attempted from thalamus or basal ganglia of horses that die within one or two days of developing clinical disease (64, 65).

Vaccines are now available for the control of EEE but are not recommended for routine use in Canada because of the extremely low prevalence of disease. However, it is recommended that horses being exported or transported to the eastern United States be vaccinated before leaving Canada.

Western Equine Encephalomyelitis

Western equine encephalomyelitis virus was first isolated in California in 1930 (7) and in Saskatchewan in 1935 (66). Like EEE, WEE is a single-stranded, enveloped, RNA virus of the genus Alphavirus, family Togaviridae (56, 67).

Western equine encephalomyelitis is believed to be maintained in a primary enzootic cycle involving *Culex tarsalis* and nestling and juvenile birds. This cycle, however, may be amplified by infection of domestic birds, wild and domestic mammals (55), and snakes (56). Although *C. tarsalis* is the principal vector of WEE virus, the virus has been isolated from several other species of mosquitoes and these may also be important in the infection cycle (67). Horses and humans are "accidental" victims of this cycle when they are fed upon by an infected mosquito, and play no role in the maintenance of this virus in nature.

Since the first Canadian isolation of WEE virus from horse brain in 1935 (66), there have been numerous epidemics of WEE in humans (68) and several epizootics in horses in western Canada (68). The most extensive horse epizootics occurred between 1935 and 1938, involving over 60,000 horses. The morbidity and mortality of the disease declined following the introduction of a horse vaccine in 1938 (69). The advent of the vaccine, however, coincided with the progressive mechanization of agriculture and a sharp reduction in the horse population. Due to the endemicity of WEE virus, small outbreaks still occur in western Canada (68). The peak incidence of disease is in July and August when mosquito activity is high (70).

The outcome of infection with WEE is similar to that described for EEE (71). Western equine encephalomyelitis virus is, however, less virulent than EEE (62) and the fatality rate of infected animals is considerably less, being 10-50% for WEE compared with 75-90% for EEE (72).

Histological lesions are also similar to those described for EEE and are typical of a nonsuppurative encephalomyelitis (73, 74).

The diagnosis of WEE in horses is often made by a combination of clinical findings, epidemiological information, and in fatal cases histopathology. Definitive laboratory diagnosis by virus isolation or by specific antibody rise or fall between paired sera is rarely accomplished. Calisher *et al* (75) showed that HI antibodies are present in the serum of many horses one or two days after the onset of clinical signs, reach a maximum five to seven days after onset of signs and thereafter decline. They suggest that a presumptive diagnosis of WEE can be made in an unvaccinated horse by demonstrating HI antibodies in a single acute phase serum sample. Neutralizing and CF antibodies rise at the end of the first week of signs and acute and convalescent sera taken 14 or more days apart can reliably demonstrate titer rises (75). Virus isolation from brain tissue is most likely to be successful if the horse died within one or two days of the onset of symptoms (73).

Vaccines available for the control of WEE in horses include: Equiloid, (Fort Dodge Labs. Inc., Fort Dodge, Iowa), Encevac 4, Encevac, and Encevac T, (Haver Lockhart, Bayvet Div., Miles Lab. Ltd., Hamilton, Ontario). Formalin-inactivated vaccines are effective and bivalent vaccines can be used; most are now produced in tissue culture. Annual vaccination is recommended and should precede the expected beginning of the "encephalitis" season by several months. In western Canada that would necessitate vaccination in the late spring.

Powassan Virus

Powassan virus is a tick-borne, single stranded RNA virus of the genus Flavivirus belonging to the family Flaviviridae (76). It was first isolated in 1958 by McLean and Donohue from the brain of a five-year-old boy, from the town of Powassan, Ontario, who died of encephalitis (77). Since that time, 19 human cases of POW encephalitis have been recorded in North America (78): eight cases in Canada, ten in New York State, and one in New Jersey.

Man appears to be a tangential host in a natural tick-rodent cycle involving small mammals such as groundhogs, squirrels, and skunks as the main reservoirs, and ticks, primarily of the *Ixodes* genera, as the main vector (8).

Powassan virus has not been isolated from any naturally infected domestic animal, although Little *et al* (79) have shown that experimental inoculation of horses with POW virus can produce a focal necrotizing meningoencephalomyelitis. In their study, two horses were inoculated intracerebrally (IC) and one intravenously (IV) with POW virus. On the eighth day postinoculation the two IC-inoculated animals developed neurological signs which included depression, head and neck tremors, slobbering, and severe ataxia. On the same day the IV-inoculated animal developed milder neurological signs. Further work in our laboratory has confirmed this pattern. A survey conducted on sera collected from 118 horses in Ontario over the 18 month period between August 1984 and January 1986 did not reveal the presence of HI antibodies to POW antigen in any animal (unpublished observations). This suggests a very low incidence of infection by POW virus in horses.

St. Louis Encephalitis Virus

St. Louis encephalitis virus is a mosquito-transmitted flavivirus of the family Flaviviridae (76). In Canada, it was first isolated in 1971 from a pool of *Culex tarsalis* mosquitoes collected in the Weyburn area of southern Saskatchewan (80). The only other isolation reported to date in Canada has been from human brain during a 1975 outbreak of SLE in Ontario (81).

St. Louis encephalitis virus is maintained in nature in an amplification cycle primarily involving birds and *Culex* species mosquitoes. Humans and most other mammals are not part

of the amplification cycle but serve as dead-end hosts (8).

Neutralizing antibodies to SLE have been recorded from horses in California, Colorado, Montana, Washington, Nevada, Massachusetts, and Texas (82, 83, 84, 85, 86). However, there have been no reported cases of naturally-occurring equine encephalomyelitis from which SLE virus has been isolated, or from which HI antibodies have been found.

In one experimental study in the USA (87), horses were inoculated IC with SLE virus and those without previous serum antibody for SLE developed clinical encephalomyelitis which was fatal in some instances. In the second study (88), horses were inoculated intranasally, subcutaneously, or intracerebrally. None of the horses developed any clinical signs, although all developed a high antibody titer. Virus was not isolated from the brain of any horse in either study.

In view of the observation by Hammon *et al* (88) that inapparent infection of horses with SLE virus occurs frequently in many widely scattered areas of the United States, an attempt was made to determine whether horses in southern Ontario had antibodies to SLE virus following the SLE outbreak in humans in 1975 and 1976 in that area. Three hundred and forty horses were tested but none had antibodies to SLE virus.

California Encephalitis Viruses

The California group viruses are mosquito-transmitted viruses of the family Bunyaviridae, comprising twelve serotypes isolated in Africa, Europe, Asia, and North and South America (56, 89). Snowshoe hare (SSH) and Jamestown Canyon (JC) are two of these serotypes which have been isolated in Canada and which are known to have the potential to cause disease in humans (8).

Snowshoe hare virus is the most widely occurring arbovirus in Canada and is maintained in an amplification cycle involving small mammals, such as snowshoe hares, and mosquitoes, primarily of the *Aedes* genus. It is transmitted transovarially in infected mosquitoes and can be maintained for many generations without requiring a vertebrate amplifying host (8).

Jamestown Canyon virus has an amplification cycle involving white-tailed deer and possibly other mammals, such as moose, and *Aedes* species mosquitoes. As with SSH virus, it is transmitted transovarially in mosquitoes (8). Jamestown Canyon virus has a more limited distribution in Canada than SSH and has been isolated from *Aedes* species mosquitoes in Alberta, Ontario, and Saskatchewan (9).

Human exposure to California serogroup viruses has been documented in six Canadian provinces, with exposure rates varying from 0.5 to 31.87% depending on the areas sampled and tests employed (9). Despite this high exposure, clinical disease rarely occurs. As of 1985 there had been only 15 symptomatic infections due to SSH and one due to JC recognized in Ontario (8).

Several serological surveys in Ontario have been conducted on horse sera and these suggest that horses also have high exposure to SSH. Sera collected in the winter of 1976 from 228 horses from southern Ontario (Guelph, Ridgetown, and Huron Park) were examined for antibodies to SSH virus (90, 91). Forty-eight (21.1%) had HI antibodies to SSH antigen, with titers ranging from 1:10 to 1:160. In the fall of 1976 and winter of 1977, sera from an additional 112 horses in five counties of southern Ontario (Elgin, Kent, Lambton, Middlesex, an Essex) were tested (91). Forty-six (41.4%) had HI antibodies to SSH antigen. More recently, 44.7% of sera collected from 114 horses throughout Ontario had HI antibodies to SSH ranging in titer from 1:10 to 1:1280 (unpublished observations).

Despite this seemingly high exposure rate of horses to SSH virus, there has been only one report of encephalitis in a horse associated with this agent (92). This occurred in a yearling hunter stallion in July 1983. The horse showed signs of acute encephalomyelitis with pyrexia, ataxia, circling, and head pressing. With conservative therapy the horse recovered within one week and had no evidence of neurological sequelae. A diagnosis was made on the basis of a fourfold or greater seroconversion to the SSH serotype of the California serogroup viruses by HI, CF, and neutralization tests.

It would thus appear that California encephalitis viruses may be responsible for some of the undiagnosed, nonfatal, acute encephalitides that are seen in horses in Ontario. Whether or not horses play a role in the actual amplification cycle of SSH virus in southern Ontario is uncertain, although possible.

Discussion

There are many causes of neurological disease in the horse and the differential diagnosis of some of these has been described elsewhere (1). A diagnosis of equine viral encephalomyelitis is made in part, by eliminating other nonviral causes such as protozoal encephalomyelitis, cervical stenotic myelopathy, and equine degenerative myelopathy.

The three principal viral agents to be considered in Canada as causes of equine encephalomyelitis are rabies, WEE, and EHVI.

The most important of these, because of its zoonotic implications, is rabies and brain tissue from every horse that dies with neurological disease should be submitted for rabies testing by FA, tissue culture and/or mouse inoculation tests. Histopathological diagnosis based on a search for Negri bodies is not adequate since in 10-15% of rabies positive cases, Negri bodies are not identified (4, 23).

Western equine encephalomyelitis must be considered the most probable clinical diagnosis in horses with neurological disease in western Canada during the mosquito season. However, one should first rule out rabies and also consider plant and fungal toxins as possible causes. A serological diagnosis can be made in animals that survive by demonstrating a fourfold or greater increase in antibody titers in paired serum samples. As it is often difficult to obtain two serum samples, a presumptive diagnosis can be made in an unvaccinated horse by demonstrating HI antibodies in a single acute phase serum sample. Virus isolation should be attempted and is more likely to be successful if the horse dies within one or two days of showing clinical signs. Brain tissue should be obtained from thalamus or basal ganglia for isolation attempts.

Equine herpesvirus type I is difficult to diagnose definitively. Horses with EHVI infection of the CNS may have a history of having been in contact with horses that aborted or had upper respiratory tract disease in the previous three months. They generally exhibit paresis and recumbency

which may resolve in seven to ten days. Often these animals will have an anamnestic response in serum antibody titers with a greater than four-fold increase between acute and convalescent samples. It may be possible to detect EHVI antibody titers in cerebrospinal fluid although early investigative work in this area has been inconclusive (95). At postmortem it is important to snap-freeze brain tissue and to attempt to isolate the virus in monolayers of susceptible cells. Histological lesions that suggest EHVI infection are vasculitis with secondary hypoxic degeneration in adjacent neural tissue. Lesions tend to be more prominent in spinal cord than in brain, and more prominent in white matter than in gray matter.

Viral isolation has proved difficult not only for EHVI but also for EEE and WEE and this may be because specialized methods of viral isolation are required or it may be due to the existence of strain peculiarities or viral neutralizing antibody masking the virus and viral antigen.

No cases of EEE have been reported in Canada since 1972 and because of its extremely low prevalence it need not be considered a likely cause of equine neurological disease. It should however, be considered if an outbreak of equine neurological disease were to occur in eastern Canada during the mosquito season, and in a horse showing neurological signs that had recently been transported into Canada from the eastern United States.

Powassan virus, although it has been shown to produce a focal non-suppurative necrotizing encephalomyelitis in experimentally inoculated horses, because of its low prevalence as indicated by a limited survey of equine serum samples, probably should not be high on the list of suspected causes of equine neurological disease.

On the other hand, serological surveys indicate that horses have a high exposure to SSH virus and this agent should be considered especially in horses that develop nonfatal acute encephalomyelitis during the summer months when the mosquito population is high.

St. Louis encephalitis virus activity has not been detected in Ontario since 1976 so it is unlikely that this agent would be responsible for causing neurological disease in horses.

There is no specific treatment for horses with viral encephalomyelitis. Some will improve with supportive therapy but others will go on to die or be euthanized for humane reasons. Vaccination as a form of control in Canada is only available for rabies, EEE, and WEE viruses. Vaccination is also available for EHVI but is not specific for the neurological form of the disease.

Acknowledgments

We gratefully acknowledge financial support from the La Foret scholarship and the Ontario Ministry of Agriculture and Food. We thank Dr. Ken Charlton of ADRI in Ottawa for providing the data on equine rabies and F. Keen and M. Blackler for their help in preparing this manuscript.

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