Hantavirus Pulmonary Syndrome in Northern Alberta, Canada: Clinical and Laboratory Findings for 19 Cases

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We reviewed the clinical and laboratory findings for 19 cases of hantavirus pulmonary syndrome (HPS) identified either serologically or by immunohistochemical testing of archival tissue at our tertiary care center. Fever (95%), cough (89%), and dyspnea (89%) were the most common presenting symptoms. The most prevalent presenting signs were respiratory abnormalities (95%) and tachycardia (84%). Common laboratory findings included thrombocytopenia (95%) and leukocytosis (79%). Elevated aspartate aminotransferase and lactate dehydrogenase levels were found in all patients tested. Intubation was required in 58% of the patients, and inotropic support was required in 53%. Our study confirms that serological responses appear early during clinical illness, making the enzyme immunoassay a useful tool for the diagnosis of acute HPS. The mortality (26%) and severity of disease that we observed among patients with HPS appear to be less than those reported elsewhere.

In fall of 1992, there was an exceptional amount of rainfall in the Four Corners region of the United States; this led to the growth of a massive deer mouse (Peromyscus maniculatus) population during the following year [1]. In May 1993, an outbreak of a mysterious disease occurred in rural New Mexico [1]. A case definition was developed by the Centers for Disease Control and Prevention (CDC) [1]. The causative agent of this disease, a hantavirus identified by the CDC by means of serological testing, was designated as “Sin Nombre” (“No Name”) virus, and the syndrome became known as hantavirus pulmonary syndrome (HPS) [1–9]. Transmission of Sin Nombre virus to humans in North America is believed to occur through exposure to deer mice or deer mice excreta [10, 11]. The mortality associated with this syndrome has been reported to be as high as 43% in the United States (as of May 1999) [12].

Before the outbreak in New Mexico occurred in 1993, there had been no reports of HPS in North America. Since the original outbreak of HPS occurred in the Four Corners region of the United States in 1993, there have been numerous reports of the disease throughout both North and South America [12–19]. In Canada, 20 cases have been reported in Alberta, 6 in British Columbia, 5 in Saskatchewan, and 1 in Manitoba (as of 1 September 1999).

We observed (a) that the cases of HPS seen in northern Alberta were associated with a mortality rate that was lower than that previously reported in the United States and (b) that the majority of cases in Canada have occurred in Alberta. This report describes the exposure history, symptoms, signs, treatment, outcome, and laboratory diagnosis of 19 cases of HPS that occurred in northern Alberta from 1989 through June 1998.

Patients and Methods

Patient population. During the period from September 1994 through June 1998, 19 cases of HPS were identified at 3 tertiary care centers in Edmonton, Alberta, Canada. Three of these cases occurred in 1989, 1990, and 1992, respectively, and were identified retrospectively with the use of archival blood and tissue samples. All cases of HPS were confirmed either by positive serology or by immunohistochemical staining of archival tissues. All cases fit the CDC case definition of HPS [1]. A chart review of the exposure history, symptoms, signs, treatment, and outcome was performed for all cases of HPS treated in Alberta, by use of a standardized database form.

Diagnostic tests. Serum samples obtained from the patients were tested against Sin Nombre nucleocapsid antigen and a normal control antigen by use of an EIA, as described elsewhere [8]. In February 1998, all but 3 of the patients’ specimens were tested at the Canadian Science Centre for Human and Animal Health, in Winnipeg, Manitoba, by use of an EIA on 1 standardized test run. Those patients whose serum specimens were not included in this test run had HPS diagnosed after December 1997. The dilutions of human serum that were used were 1:100, 1:400, 1:1600, and 1:6400. The cutoff point for a positive test was an optical density of 0.1 when compared to background. The serum titer was determined to be the highest dilution at which a positive result was obtained. Formalin-fixed tissue specimens from 2 patients were tested by means of immunohistochemical staining (testing was done by Dr. S. Zaki, CDC, Atlanta) [8]. Blood clots and tissues from both a...
representative seropositive deer mouse and a patient with HPS were used for reverse transcriptase–PCR amplification of a portion of the hantavirus G1 gene, with use of primers described by Johnson et al. [20].

Statistical analysis. A 2-tailed nonparametric 1-sample t test (\( \alpha = .05 \)), done with the use of SPSS software, version 9.0 (SPSS, Chicago), was performed to compare the number of days from onset of symptoms to presentation in the patients who survived and in those who died.

Results

The medical records of all 19 patients were available for this study. The median age of the patients was 40 years (range, 15–65 years). Eleven (58%) of the patients were men. The median time from onset of symptoms to presentation was 5 days (range, 2–19 days). The majority of cases of HPS occurred in the spring (figure 1). Ten (53%) of the 19 cases presented between 1 June 1997 and 19 June 1998.

All of the patients were treated at our 3 tertiary care centers. Eighteen of the 19 patients were from rural Alberta, and 1 patient, who was working in Fort St. John, British Columbia, was from Saskatchewan (figure 2). All but one of the patients had a known exposure to mice or mice excreta in the 6 weeks prior to the onset of illness. The one patient who did not have an identified history of exposure to mice had a pet gerbil at home. A blood sample obtained from the gerbil was sent for serological testing and was found to be negative for evidence of hantavirus infection.

Symptoms and signs. In a retrospective study, it is difficult to ascertain whether the physician who obtained the history asked about specific symptoms, since negative findings are sometimes not recorded. We assume that a search for the symptoms listed below was done for all critically ill patients. The most common symptoms at the time of presentation were fever (95%), cough (89%), and dyspnea (89%). In 7 (41%) of the 17 patients, the cough was nonproductive. Other frequently reported symptoms included nausea (74%), chills (63%), and vomiting (58%). Headache was reported by 12 patients (63%), of whom 4 had a lumbar puncture performed as part of their initial workup. The occurrence of rhinorrhea and sore throat, symptoms that may not have been consistently sought, was sometimes not recorded. We assume that a search for the symptoms, since negative findings are not recorded. We assume that a search for the symptoms listed below was done for all critically ill patients. The most common symptoms at the time of presentation were fever (95%), cough (89%), and dyspnea (89%). In 7 (41%) of the 17 patients, the cough was nonproductive. Other frequently reported symptoms included nausea (74%), chills (63%), and vomiting (58%). Headache was reported by 12 patients (63%), of whom 4 had a lumbar puncture performed as part of their initial workup. The occurrence of rhinorrhea and sore throat, symptoms that may not have been consistently sought, was sometimes not recorded. We assume that a search for the symptoms listed below was done for all critically ill patients.

The most prevalent presenting signs were respiratory abnormalities (95%), tachypnea (84%), and tachycardia (84%). Hypotension at presentation, which was defined by a value of <100 mm Hg systolic pressure, was noted in only 32% of patients. Abnormalities on chest radiography were seen in 18 (95%) of 19 patients at the time of presentation. All of these patients had interstitial infiltrates, and 5 (26%) of 18 patients had pleural effusions.

Laboratory values. Common hematological abnormalities that were seen during the clinical course of HPS included thrombocytopenia (95%) and leukocytosis (79%). The trough median platelet count was 52 x 10^3 cells/L (range, 193–9 x 10^3 cells/L), and the peak median WBC count was 18.5 x 10^3 cells/L (range, 5.7–59.2 x 10^3 cells/L). Increased hemoglobin and hematocrit were seen in 5 (26%) of the 19 patients. The partial thromboplastin time was increased in 16 (89%) of 18 patients. Five (45%) of the 11 patients who had a peripheral blood smear prepared, with or without bone marrow aspiration, had atypical lymphocytes seen by a hematopathologist.

The other laboratory abnormalities included elevated levels of aspartate aminotransferase in 17 patients and elevated levels of lactate dehydrogenase in 15 patients. The peak median value was 124 IU/L (range, 57–23,120 IU/L) for aspartate aminotransferase and 657 IU/L (range, 279–20,200 IU/L) for lactate dehydrogenase. Thirteen (93%) of 14 patients had hypoalbuminemia. An increased creatinine kinase concentration was seen in 6 (43%) of 14 patients. The creatinine level was elevated in 58% of patients. For 6 patients, the median arterial partial pressure of O₂ on room air at the time of admission was 46.5 mm Hg (range, 39–51 mm Hg). All of the patients required supplemental oxygen at the time of admission.

The plasma lactate level was elevated in 5 (50%) of 10 patients, and the venous bicarbonate level was low in 15 (79%) of 19 patients during the hospital days prior to and during the first week in the intensive care unit. The peak median level of plasma lactate was 2.7 mM/L (range, 1.0–20.4 mM/L), and the trough median level for venous bicarbonate was 18 mM/L (range, 8.2–25 mM/L) during this time.

Clinical outcome. Five (26%) of the 19 patients died. One (33%) of the 3 patients who had HPS diagnosed retrospectively before 1994 and 4 (25%) of the 16 patients who had HPS diagnosed after 1994 did not survive. For the 10 cases diagnosed from 1997 through 1998, the mortality rate was 30%.

The median time from the onset of symptoms to presentation was 6.5 days (range, 2–19 days) for the patients who survived.

Figure 1. Seasonality of cases of hantavirus pulmonary syndrome (HPS) in northern Alberta, Canada (as of December 1998). Each pattern represents a year in which at least one case of HPS was diagnosed.
and 3 days (range, 2–6 days) for those who died. With regard
to the time from onset of symptoms to presentation, there was
no significant difference between the survivors and those who
died.

Intubation was required for 11 (58%) of 19 patients. For
those patients who survived, the median time from onset of
symptoms to intubation was 6.5 days (range, 4–21 days); for
those who died, it was 4 days (range, 2–6 days). The median
duration of intubation was 6 days (range, 2–70 days) for those
who survived and 1 day (range, 1–4 days) for those who died.
Ten (71%) of the 14 patients who survived had a mean arterial
pressure (MAP) of ≤70 mm Hg recorded during the clinical
course of HPS, and 5 (50%) of these 10 patients required inotrop support. All patients who died had a MAP of ≤70 mm
Hg during the clinical course of HPS and required inotropic
support.

The median duration of hospitalization was 9 days (range,
6–85 days) for those who survived, and for those who died, the
median interval from presentation to death was 2 days (range,
1–5 days). Two (14%) of the 14 surviving patients were treated
with ribavirin, and 5 patients (26%) were enrolled in a ran-
domized clinical trial of ribavirin versus placebo.

**Diagnostic testing.** Blood samples for serological testing
were collected from all of the patients (table 1). A positive IgM
response was seen in 17 (89%) of 19 patients at the time of the
first serological testing, at a median of 7 days (range, 2–21 days)
after the onset of symptoms. One of the patients who was
initially seronegative had a positive IgM response when retested
14 days after the onset of symptoms, and the other (for whom
no further serological results were available) had the diagnosis
confirmed by immunohistochemical staining of tissue. Blood
samples from this patient had been collected in 1989. The long
period of storage may account for the lack of IgM reactivity.

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a Number of days from onset of symptoms to serology.
b HPS was diagnosed in this patient by means of immunohistochemical stain-
ing of archival tissues. A blood sample had been collected from this patient in
1989, which may account for the lack of IgM reactivity.

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Figure 2. Geographic locations of cases of hantavirus pulmonary syndrome (HPS) in northern Alberta, Canada. One patient, who was referred
to our tertiary care center from Fort St. John, British Columbia, was a resident of Saskatchewan. Squares, cities; circles, cases of HPS.
The IgM response was positive in 14 (88%) of 16 patients who had their blood collected within 10 days of the onset of symptoms, and it lasted as long as 34 days in 1 patient. After this point, the IgM response begins to decline. In 10 (53%) of 19 patients, an IgM and IgG response was seen during the first serological test. During subsequent testing, which was done at a median of 25 days (range, 16–31 days) after the onset of symptoms, a further 4 (44%) of 9 patients had a Sin Nombre–specific IgG response detected. Four years after development of acute HPS, 1 patient had an IgG titer of 1:400 to Sin Nombre virus.

Sequence analysis of hantavirus G1 amplicons generated from both a patient with HPS (patient 8; table 1) and a seropositive deer mouse from northern Alberta seems to show that these hantaviruses were genotypically related to each other (figure 3). Although genotypically distinct from Sin Nombre and Convict Creek 107 strains, the northern Alberta hantaviruses appear to be more closely related to these strains than to eastern North American hantaviruses, such as the New York or Black Creek Canal viruses (figure 3).

Discussion

During the period from June 1997 through June 1998, 10 cases of HPS were diagnosed at our centers; this is a greater number of cases than was reported in the area during the period from 1989 through 1996. As previously discussed with regard to the Four Corners outbreak, El Niño weather effects seemed to boost the deer mouse population [1]. A similar population explosion may have occurred among deer mice in Alberta during the summer of 1997, resulting in increased exposure of humans to mice. In 1997, most of the cases of HPS occurred during the fall, when the mice may have been seeking shelter indoors.

The age range of the patients varied widely in our study, with the youngest patient being 15 years old. This is similar to findings from studies in the United States, where, as of 4 February 2000, there have been no confirmed cases involving children aged <10 years old (www.cdc.gov/ncidod/diseases/hanta/hps/noframes/caseinfo.htm). Why children do not contract the illness, however, is not entirely understood. One hypothesis is that HPS is immunopathological in nature and that repeated exposure to Sin Nombre virus is required to cause disease.

Analysis of the geographic locations of the cases of HPS on a map of Alberta reveals that they fit into a belt-like pattern in the midregion of the province (figure 2). Recent studies have suggested that multiple environmental factors may play a part in the increase in the number of deer mice and in subsequent increases in the number of cases of HPS [22]. Most of the exposures in our study involved the inhalation of aerosols from areas contaminated with mouse excreta.

The symptoms and signs observed at presentation in this study are similar to those reported previously, except for a lower incidence of markers of severe disease, such as hypotension, which was noted in 32% of patients, compared with 50% of patients in an earlier study [4]. Only 2 patients had rhinorrhea and a sore throat, a finding demonstrating that these symptoms are useful in differentiating HPS from disease caused by influenza [22]. In contrast to a previous study of symptoms seen with HPS, many patients in our series had cough as a presenting symptom, suggesting that lack of cough was not useful in differentiating HPS from other causes of pneumonia [23].

The laboratory results seen in this series of cases are similar to those reported elsewhere, with thrombocytopenia, leukocytosis with a left shift, and an increase in lactate dehydrogenase level being common [1–9]. However, the patients in our study did not have the same incidence of hemoconcentration, which is another marker of severe disease. A recent study also showed that the finding by hematopathologists of an increase in atypical lymphocytes in peripheral blood smears is useful for the diagnosis of HPS; this observation was made for 45% of our patients [1]. In this study, the aspartate aminotransferase level was elevated in 100% of the patients, a finding that suggests that a normal result makes a diagnosis of HPS far less likely.

A study of the first 100 cases found in the United States showed that 65 (84%) of 77 patients required intubation, a proportion that is higher than the 11 (58%) of 19 patients who required intubation in our study [5]. When hypotension was recorded during the clinical course of HPS, 79% of patients had a MAP of <70 mm Hg, and 53% required inotropic support. Data on hypotension during the clinical course of HPS have not been given in previous study reports.

Our study confirms that serological responses appear early during clinical illness, making the EIA a useful tool for the diagnosis of acute HPS. The IgM serology for 1 of the patients was negative on day 6 of the illness but became positive on day 14; this finding emphasizes that if there is a clinical sus-
picion of HPS, serological testing should be repeated. The IgG antibodies to Sin Nombre virus seem to persist for a long time, as was illustrated by the patient who had an IgG titer of 1:400 4 years after acute HPS developed.

A recent study indicated that the Sin Nombre–like viruses found in Alberta deer mice were genotypically distinct from the Sin Nombre virus strains in the southwestern United States [24]. However, this study also showed that western North American Sin Nombre–like viruses seemed, at the genotypic level, to be more related to each other than to strains found in eastern regions of the continent. Our findings are consistent with this observation. Northern Alberta viral strains both from a patient with HPS and a seropositive deer mouse also showed a closer genetic relationship to western Sin Nombre viruses than to such viruses as the New York or Black Creek Canal viruses.

In our series, we observed a lower incidence of signs of severe disease at admission, a lower incidence of intubation, and a lower mortality (26%) than those observed by other investigators. The case-fatality rate of 25% seen since 1994 in our series of patients can be compared with the case-fatality rate of 34% reported from the United States since 1994 (as of 4 February 2000); however, because of the small number of patients in our study, this difference may not be significant (www.cdc.gov/ncidod/diseases/hanta/hps/noframes/caseinfo.htm). The low case-fatality rate observed may be the result of earlier detection of infection, aggressive intensive care unit management, the viral inoculum to which the patients were exposed, the genetic predisposition of the patients, or the presence of a variant of the virus in northern Alberta. Further genetic analyses of Sin Nombre isolates from our region and from other regions would be useful for differentiating between these possibilities.

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References