ORIGINALARTICLE

Calgary Experience with West Nile Virus Neurological Syndrome During the Late Summer of 2003

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ABSTRACT: ***Background:*** Between August 25 and September 25, 2003 seven patients with West Nile virus neurological manifestations were identified through the hospital neurology consultation services in Calgary, Alberta, Canada. Three of the seven patients were treated with interferon alpha-2b (IFN alpha-2b). In this report we document the clinical characteristics of these seven cases. ***Methods:*** Clinical and laboratory information was obtained from a retrospective review of patient hospital and clinic charts. Patients were included if they had serological evidence of West Nile virus infection and had clinical evidence of aseptic meningitis, encephalomyelitis, cerebellar syndrome or motor neuronopathy. Three patients received a treatment course of three million units IFN alpha-2b, administered by subcutaneous injection once per day for 14 days. ***Results:*** Four patients had cerebellar signs without change in consciousness, two had both encephalitis and neuromuscular weakness, and one patient had focal lower motor neuron arm weakness. The mean age was 52 (range 24 – 73). All patients had flu-like illness and fever as presenting symptoms and six had severe headaches. Two patients were immunocompromised prior to infection. Two patients with cerebellar signs (one with opsoclonus-myoclonus) improved spontaneously and exhibited only mild residual deficits on discharge. The other two patients with cerebellar findings developed brainstem involvement, one coinciding with and one subsequent to the cerebellar symptoms. Within one week of treatment with IFN alpha-2b these latter two patients showed marked improvement. One patient with encephalitis and neuromuscular weakness, was treated with IFN alpha-2b and subsequently recovered. ***Interpretation:*** In this case review of seven patients, multiple neurological symptoms occurred in each patient and the neurological presentation was varied. Four patients had predominant cerebellar findings and one patient had opsoclonus-myoclonus, not previously reported. The marked improvement in three patients who received IFN alpha-2b raises preliminary optimism towards this potential treatment.

RÉSUMÉ: Syndrome neurologique dû au virus du Nil occidental en fin d'été 2003 à Calgary. ***Introduction:*** Entre le 25 août et le 25 septembre 2003, sept patients ont consulté à Calgary, Alberta, Canada, pour des manifestations neurologiques dues au virus du Nil occidental. Trois des sept patients ont été traités par l'interféron alpha-2b (IFN alpha-2b). Nous décrivons les caractéristiques cliniques observées chez ces patients. ***Méthodes:*** Il s'agit d'une revue rétrospective de dossiers cliniques et hospitaliers, incluant les épreuves de laboratoire, l'analyse du liquide céphalo-rachidien (LCR), les résultats d'examens électrodiagnostiques et d'imagerie le cas échéant. Les patients chez qui la sérologie était positive pour une infection par le VNO et qui avaient des manifestations de méningite aseptique, d'encéphalomyélite, de syndrome cérébelleux ou de neuronopathie motrice ont été inclus dans l'étude. Trois patients ont reçu une série d'injections sous-cutanées quotidiennes de trois millions d'unités d'IFN alpha-2b pendant 14 jours. ***Résultats:*** Dans cette série de patients, quatre patients avaient des signes cérébelleux sans modification de l'état de conscience, deux avaient une encéphalite accompagnée faiblesse neuromusculaire et un patient avait une faiblesse au niveau du bras due à une atteinte focale du neurone moteur périphérique. L'âge moyen des patients était de 52 ans (écart de 24 à 73 ans). Tous les patients présentaient des symptômes grippaux et de la fièvre à la consultation initiale et six avaient des maux de tête sévères. Deux patients étaient immunocompromis avant l'infection. Deux patients ayant des signes cérébelleux, dont un ayant de l'opsoclonie-myoclonie, se sont améliorés spontanément et ne présentaient que de légers déficits résiduels à la sortie de l'hôpital. Deux autres patients ayant des manifestations cérébelleuses ont développé une atteinte du tronc cérébral, concomitante à l'apparition des symptômes cérébelleux chez l'un et postérieure chez l'autre. L'état de ces deux patients s'est amélioré considérablement après une semaine de traitement par l'IFN alpha-2b. Un patient présentant une encéphalite et de la faiblesse neuromusculaire a récupéré suite au traitement par l'IFN alpha-2b. ***Conclusion:*** Les sept cas inclus dans cette revue ont tous présenté de multiples symptômes neurologiques avec un tableau neurologique variable d'un patient à l'autre au moment de la consultation initiale. Chez quatre patients, les signes cérébelleux prédominaient et un patient avait de l'opsoclonie-myoclonie, ce qui n'a jamais été rapporté. L'amélioration importante observée chez les trois patients de cette série, qui ont reçu de l'IFN alpha-2b, permet de croire que ce traitement pourrait être efficace.

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The West Nile virus (WNV) is an arthropod borne virus that is serologically classified amongst the group of Japanese encephalitis flaviviruses and it is antigenically similar to the Kunjin, Murray Valley, and St. Louis encephalitis viruses. It is transmitted to humans through the *Culex* mosquito and typical reservoir species include a variety of wild and domestic birds, horses and other mammals.[1] Transmission through blood transfusion,[2] organ donation,[3] pregnancy,[4] lactation[5] and needle stick injury[6] has been previously described. West Nile virus infection was first isolated from humans in Uganda in 1937.[7] It has subsequently been described in epidemic forms throughout Africa, Asia and the Middle East.[1,8]

The first cases of WNV infection in North America occurred in 1999 in the New York City outbreak. Between August 2nd and September 24th 1999, there were 59 cases of patients with encephalitis or meningitis who tested positive for WNV infection.[9] Over the following three years the virus spread throughout the eastern USA and reached Canada in 2002. Ontario was the first province to encounter WNVinfection in the summer of 2002 when an epidemic of 62 cases with neurological manifestations occurred.[10] Other than two travel-related cases of human illness in 2002, the first sign of the virus in the province of Alberta appeared July 9, 2003 when a magpie found near Camrose, Alberta tested positive for WNV.[11] Entomologists tracking the migration of arthropod vectors have already illustrated the WNV encroaching upon North America's western shores.[12]

Most cases of WNVinfection are subclinical, with symptoms of West Nile Fever occurring in approximately 20 percent of people who are infected.[13] West Nile virus fever usually begins with sudden onset febrile illness, accompanied by headaches, myalgias and gastrointestinal symptoms such as nausea and vomiting. Symptoms usually last one week followed by a prolonged period of fatigue.[14] A range of neurological syndromes

Table 1: Alberta Health Region frequency data for the 270 WNV patients reported in 2003.

REGION / CITY (Provincial Location)	Number of patients (%). WNV No Symptoms	WNV Fever	WNV Neurological Syndrome
Palliser / Medicine Hat (Southeast)	2	107 (49)	19 (40)
Chinook / Lethbridge (Southwest)	0	30 (14)	7 (15)
Calgary / Calgary (Southcentral)	1	27 (12)	8 (17)
David Thompson / Red Deer (Western)	0	21 (10)	6 (13)
Capital / Edmonton (Central)	0	16 (7)	4 (8)
East Central / Lloydminister (Eastern)	0	16 (7)	2 (4)
Aspen / Lac la Biche (Northcentral)	0	0 (0)	2 (4)
Northern Lights / High Level (Northeast)	0	2 (1)	0 (0)
Peace Country / Peace River (West Central)	0	0 (0)	0 (0)
Provincial Totals	**3**	**219**	**48**

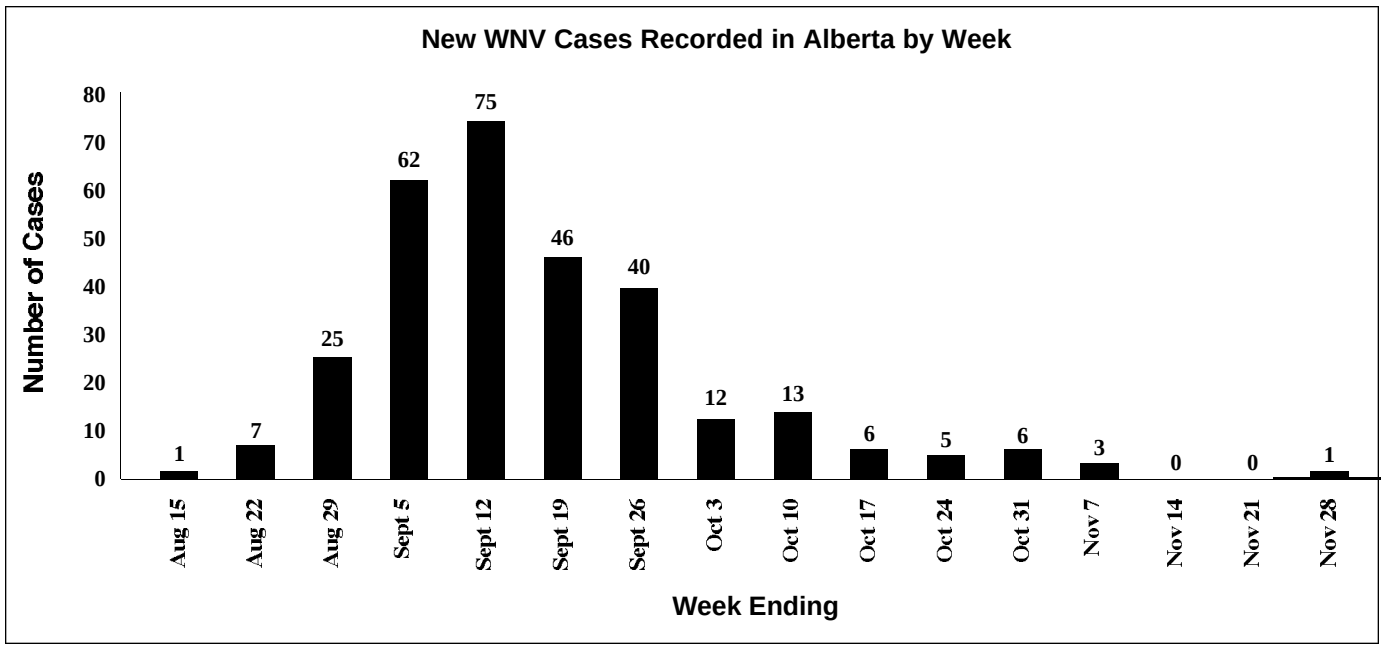

Figure 1: *Frequency of new WNVhuman patients recorded in the province of Alberta from August 15, 2003 to November 28, 2003.*

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Table 2: Characteristics of 7 patients with WNV Neurological syndrome from August 25th until Sept 25th 2003.

Patient	# 1	# 2	# 3	# 4	# 5	# 6	# 7
Age	54	39	24	73	71	56	50
Gender	Female	Female	Female	Female	Male	Male	Male
Co-morbid medical condition and past medical history	Hypertension Diabetes mellitus type two, on oral hypoglycemics Prior cholecystectomy Prior hysterectomy	Migraine with aura	Migraine with sensory symptoms Prior Caesarian-section and intrauterine device insertion	Hypertension Diabetes mellitus Type 2, diet controlled Benign paroxysmal positional vertigo	Previous cardiac bypass Diabetes Congestive heart failure Hypothyroidism Atrial fibrillation requiring electrical cardioversion	None	Childhood strabismus Tonsillectomy
Immuno-compromising condition	Breast cancer treated with 4 chemotherapy cycles	None	None	Untreated chronic lymphocytic leukemia for 5 years	None	None	None
Symptoms and signs at presentation	Five-day history of fever, chills, rigors, nausea, vomiting and diarrhea.	One week history of progressing headache, fever, rash, nausea and vomiting	One week history of headache, rash and fever	One week history of arthralgia, myalgia and fever	Two week history of fever, diaphoresis, fatigue and headache	Headache, fevers, fatigue, diarrhea and malaise for three days	Two week history of fever, chills, headache, rash, lightheadedness and anorexia
Activity prior to illness	Unknown	Vacationing on a lake SE Alberta	Two weeks working in the woods Eastern Alberta	Camping near Alberta-US border two weeks prior. Multiple visits to farms	Frequently outdoors gardening or doing carpentry in Southern Alberta	Often outside during the day	Grain farmer, outside on fields every day
Rash during prodrome	No	Yes	Yes	No	Yes	No	Yes
Recollection of mosquito bites	No	Yes	Yes	Yes	Yes	No	No

can manifest with WNV infection including encephalitis, meningitis, neuromuscular weakness and cerebellar ataxia and reports of such cases have already been published.[9,10,14-17] Serology studies from the New York City outbreak identified that approximately one out of 140 patients with infection by WNV will develop meningoencephalitis, although this value is age related and ranges from 1/50 in patients over the age of 65 to 1/300 in patients less than 65 years.[9] Despite small numbers, both the New York City and Ontario study demonstrated significant morbidity and mortality from the neurological syndrome of WNV infection.[9,10] To date, the treatment for WNV infection is prevention and supportive treatment once symptoms arise. Two potentially therapeutic agents, ribavirin and interferon alpha-2b (IFN alpha-2b) have been shown to eradicate the virus *in vitro*[18,19] and a clinical trial using IFN alpha-2b is currently underway.[20,21]

In the early summer of 2003, the first mosquitoes harboring active WNV infection were detected in traps and larval reservoirs in the Alberta region.[11] Dead birds and horses infected with WNV had been identified along the southeastern border of the province.[12] Following the discovery of the first WNV bird case, the Alberta Health and Wellness surveillance program tested 1483 birds. Between July 9, 2003 and October 20, 2003, 233 infected birds were identified in all regions except the northwest quadrant of the province.[22] New WNV human cases were recorded by week from August 15, 2003 through November 28, 2003. The maximum frequency of 75 cases in Alberta was registered during the week ending September 12, 2003 (Figure 1). In the Calgary region 8/36 cases were diagnosed with WNV neurological syndrome in 2003 (Table 1). The current article reviews seven of the eight cases of WNVneurological syndrome diagnosed in Calgary. It describes how these cases relate and differ from prior North American experience with WNV neurological syndrome and reports three cases of successful treatment with IFN alpha-2b.

METHODS

Patients with neurological manifestations of WNV infection between August 25th and September 25th, 2003 were identified through the neurology consultation services at Peter Lougheed Centre and Foothills Hospital, Calgary, Alberta, Canada. Information was obtained from a retrospective review of patient hospital and clinic charts (if follow-up appointments occurred), and through interviewing patients and their families. Laboratory tests, cerebrospinal fluid analysis (CSF) analysis, electrodiagnostic tests results (electroencephalogram (EEG), electromyography (EMG), and nerve conduction studies) and neuroimaging results were obtained if performed.

Cerebrospinal fluid analysis and serological studies were performed by the Alberta Provincial Laboratory for Public Health (Microbiology) for evidence of WNV infection. Only patients with laboratory evidence of acute WNV infection were included in this series. It has been previously shown that immunocompromised patients can have a very delayed antibody response to WN viremia with no response detected for periods up to 52 days.[9,10] Also, a four-fold increase in WNV specific neutralizing antibody titre in repeat samples during acute infection has been suggested as confirmation of the diagnosis and is one of the definitions for WNV infection used in the United States.[22] Therefore, serial WNV serology for IgM titres was performed when possible.

West Nile IgM serology was performed using newly-available commercial enzyme-immunoassay (EIA) kits, (Focus Technologies, Cypress, CA, and PanBio, Brisbane, Australia), as per the manufacturer's instructions. Two independent assays were done on each serum sample using separate kits. Positive results were reported when these assay tests were concordant. Hemagglutination inhibition titres (HI) were performed by standard methods[23,24] using antigen provided by Dr. Mike Drebot, National Microbiology Laboratory, Winnipeg. Polymerase chain reaction (PCR) was performed on 250 uL of CSF using a commercial kit (Artus, San Francisco CA) following RNAextraction (Qiagen, Cambridge MA). One mLof plasma was used for detection of viremia, with RNA extraction by the Boom method[25] followed by amplification using a published nucleic acid sequence-based amplification (NASBA) method.[26] All positive nucleic acid tests were confirmed by the second amplification method targeting an entirely separate segment of the viral genome, before reporting.

Interferon alpha-2b was offered as a treatment option to three of the seven patients and was accepted by all three. This was administered according to the New York Hospital Queens Clinical Trial Protocol.[20,21] The treatment protocol, a sample consent form, a case report form, instructions for pharmacists, and contact information is available on the New York Hospital Queens' website.[20,21] The rationale for initiating this treatment was based on previously reported laboratory research that has demonstrated the effectiveness of IFN alpha- 2b in inhibiting the growth of WNV in infected cell cultures.[18,19] In a collaborative study by New York Hospital Queens, the Connecticut Agricultural Research Laboratory, and Schering-Plough Research Institute, both ribavirin and IFN alpha-2b were tested against a WNV infected cell culture.[18,19] IFN alpha-2b inhibited viral replication at a relatively low concentration (5.9 units/ml) when applied after infection of green monkey kidney cells with WNV.[19] A dose of 3 million units of IFN alpha-2b in humans provides serum levels of 10 units/ml after eight hours, and daily doses of 3 million units yield serum levels of 20 – 30 units/ml, which are above the concentrations required for *in vitro* efficacy against WNV.[27] Therefore, in the current case report, a treatment course of 3 million units of IFN alpha-2b, administered by subcutaneous injection once per day for 14 days was used, as recommended in the New York Protocol.[20] Side effects to this treatment are relatively benign and include fever, chills, weakness, neutropenia and elevated liver enzymes.[20] Routine laboratory investigations including daily hematological tests and liver enzyme tests every other day were performed to monitor potential side effects. All patients who began treatment were given the option to terminate treatment at any time. The remaining four patients were given supportive treatment only. All patients were followed for a minimum of two weeks from symptom onset and the short-term outcome of WNV neurological syndrome was evaluated.

These cases are the first reported cases of WNV in the Calgary region and, therefore, WNV screening in patients with fever and meningoencephalitis, neuromuscular weakness or cerebellar ataxia was not routinely performed. Serologic testing

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Table 3: Clinical Characteristics of Seven Patients with WNV Neurological Syndromes Diagnosed in the Calgary area in 2003.

CHARACTERISTIC	NUMBER OF PATIENTS (%)
Syndromes	
Cerebellar	
with weakness	2
without weakness	2
Encephalitis with weakness	2
Poliomyelitis-like syndrome	1
Signs and symptoms	
Fever (temperature >37.8 C)	7 (100)
Fatigue	6 (85)
Headache	5 (72)
Weakness	5 (72)
Gait Ataxia	4 (57)
Rash	4 (57)
Nausea/Vomiting	4 (57)
Diarrhea	3 (43)
Myalgia	3 (43)
Tremor	3 (43)
Slurred Speech	3 (43)
Altered Mental Status	2 (28)
Facial Weakness	2 (28)
Opsoclonus/myoclonus	1 (14)

for WNVwas only performed in patients when there was clinical suspicion of WNV neurologic syndrome. Therefore, the complete extent of WNV neurological activity in the Calgary region remains currently unknown (Table 1). The current study provides a preliminary analysis of disease presentation, course, severity and outcome in seven cases identified during a one month time period.

Case descriptions

Below are brief synopses of the patients with WNV neurological syndrome from August 25th until Sept 25th 2003. Patient demographics and background characteristics can be found in Table 2. Clinical characteristics and CSF findings are listed in Tables 3 and 4.

Patient #1

A 54-year-old female presented with a history of fever, chills, nausea, vomiting and diarrhea for five days. She had been diagnosed with breast cancer six months before this hospital admission and had received the fourth chemotherapy cycle of cyclophosphamide/adriamycin one month prior to this admission. She developed anemia, and required admission to a peripheral hospital for blood transfusions and treatment of continuing symptoms. Upon hospitalization, she was lucid and moving all four limbs. Over 72 hours she developed a flaccid, areflexic quadriplegia and respiratory distress requiring intubation and transfer to the intensive care unit (ICU) in a Calgary hospital. Electrodiagnostic studies showed reduction in amplitude of motor responses in arms and legs with latency and conduction velocity, whereas sensory responses were normal. The needle EMG studies demonstrated early denervation changes in all four extremities. Her level of response to verbal and tactile stimuli deteriorated to the point where she rarely and inconsistently opened her eyes to command. Brainstem reflexes were impaired: corneal reflexes were weak, extraocular movements were absent and unresponsive to oculocephalic maneuvers. An MRI of the brain identified areas of T2/FLAIR hyperintensity within the posteromedial temporal lobes bilaterally, the splenium of the corpus callosum, the substantia nigra of the midbrain, posterior mid portion of pons and midline of the medulla. (Figure 2). An EEG showed mild to moderate diffuse encephalopathy with no evidence of epileptiform activity. Serum was positive for WNV IgM antibodies. Plasma and CSF were positive for WNVRNAby NASBAand PCR, respectively. She remained unresponsive to verbal and tactile stimuli, intubated and ventilated in the ICU with persistent flaccid paralysis for two weeks. She continued to deteriorate and died.

Table 4: CSF analysis of WNV patients

Patient:	Cell Count	Differential			Glucose		Protein	RBCs	CSF	CSF	Plasma	Serum
	(x 10^6)/L	Ntp	Lym	Mc	Serum	CSF(mmol)	(gm/L)	(x 10^6)/L	Cultures	PCR	PCR	IgM
# 1	10.5		77%		N	N	0.92		-ve	+ve	+ve	+ve
# 2	31.0	4%	95%	1%	N	3.5	0.44	358	-ve	-ve	-ve	+ve
# 3	20.5	57	42	1	N	3.0	0.38	12.8	-ve	+ve		+ve
# 4	143.3	63%	37%	0	N	3.5	0.65	1.1	-ve	-ve		+ve
# 5												+ve
# 6									-ve	-ve	-ve	+ve
# 7	1193.0	86%	12%	2%	6.2	3.3	1.82	19	-ve	-ve	+ve	+ve

Ntp=neutrophils, Lym=lymphocyte, Mc=monocyte, RBCs=red blood cell counts, PCR=polymerase chain reaction, IgM=immunoglobulin M

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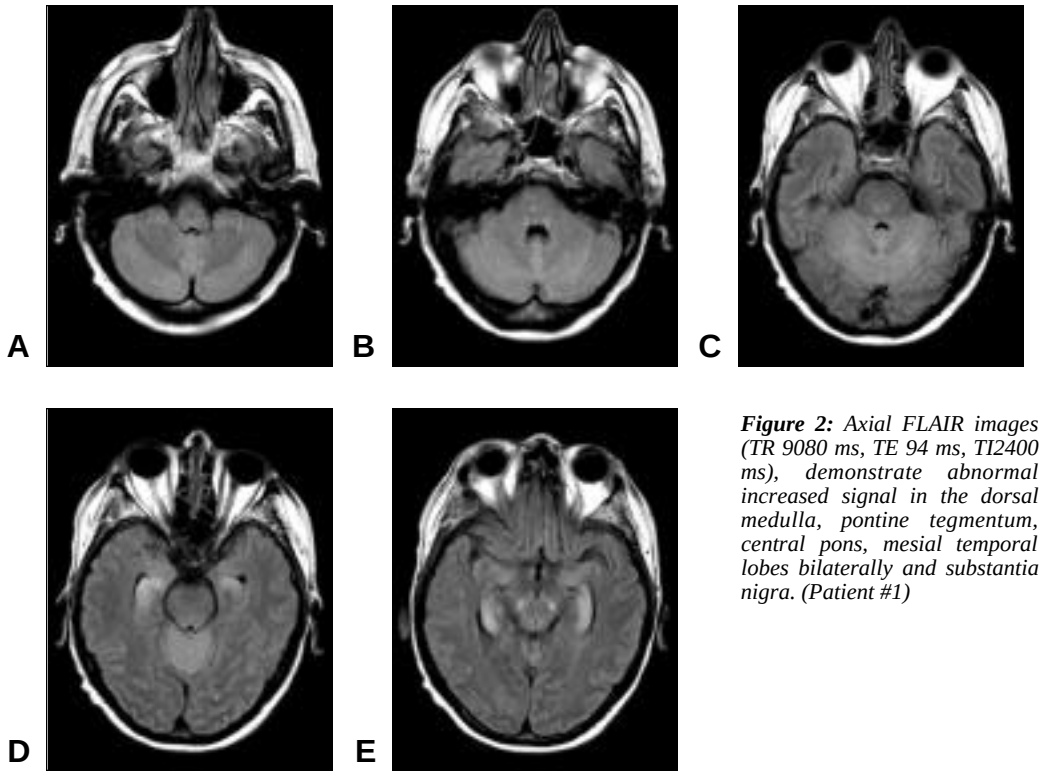

Figure 2: *Axial FLAIR images (TR 9080 ms, TE 94 ms, TI2400 ms), demonstrate abnormal increased signal in the dorsal medulla, pontine tegmentum, central pons, mesial temporal lobes bilaterally and substantia nigra. (Patient #1)*

Patient #2

This 39-year-old woman presented with a one-week history of progressive headache, severe nausea and vomiting, fever and imbalance. She had a generalized erythematous, nontender and nonpruritic skin rash that developed on her first day of symptoms. The rash persisted for 48 hours and spontaneously resolved. Three days later, the patient noticed irregular jerky eye movements, marked unsteadiness on her feet and oscillopsia. She did not experience weakness or other neurological symptoms. Physical examination revealed opsoclonus, mild left facial palsy, bilateral mild intention tremor of the upper extremities, left dysdiadokokinesis and left arm myoclonus. Her gait was ataxic and wide based. Brain MRI was normal. Serum was positive for WNV IgM and HI titre was stable 1:320. Over the course of one week this patient spontaneously gradually improved. On discharge, her neurological examination demonstrated resolution of opsoclonus-myoclonus, and mild ataxia persisted. When seen in follow-up, she complained of persisting nausea, moderate fatigue and anxiety.

Patient #3

This 24-year-old female presented with a one-week history of profound fatigue and an erythematous maculopapular rash on her trunk and extremities. Two days later she developed a severe bifrontal headache, stiff neck and a low-grade fever. She presented to the hospital one week later because of increasing fever, diarrhea, worsening occipital headache and neck pain, slurred speech and an unsteady gait. Tremor was present when reaching for objects with the right hand. The rash was improving but persisted on presentation. On examination she was febrile, her neck was supple, but posterior occipital lymphadenopathy was present. Neurological examination revealed mild dysarthria, mild right gaze nystagmus, slight left intention tremor and normal heel-shin coordination. Gait was wide based and severely ataxic. The remainder of the neurological and physical examination was normal. Brain MRI was normal. Serum was positive for WNV IgM. This patient remained in hospital for 48 hours, after which she left against medical advice. Upon discharge she was afebrile, her tremor and nystagmus had resolved completely, and her gait had improved but remained residually wide based. She did not pursue any further neurological follow-up.

Patient #4

This 73-year-old female presented with a one-week history of myalgia, arthralgia and fever. She had a five-year history of untreated chronic lymphocytic leukemia (WBC 40.0 10 E9/L, lymphocytes 38.5 10 E9/L). Four days later she developed weakness in her lower extremities, more prominent proximally and worse on the left side. She had been initially assessed in a peripheral hospital and was treated empirically with antibiotics. Over 48 hours she developed increasing confusion, disorientation and deterioration in motor function. She was transferred to Foothills hospital obtunded, with intact brainstem reflexes and able to withdraw all four limbs to pain. Left leg weakness was noted with hip flexion Medical Research Council grade of 4/5 and foot dorsiflexion grade of 3/5. Nerve conduction studies were performed which showed normal motor nerve conduction velocities with minimal change in F wave latencies. Sensory responses were normal. The needle EMG changes recorded in the left vastus lateralis and left tibialis anterior were consistent

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with early mild denervation. An MRI of the brain with T2-weighted images demonstrated mild increased signal within the right thalamus and minimal increased signal bilaterally adjacent to the globus pallidus and internal capsule. A few small scattered subcortical white matter signal hyperintensities were reported as nonspecific and possibly age related. An ill-defined T2 hyperintensity was seen in the lower thoracic cord extending from T10 to conus level. Repeat MRI of the thoracic spinal cord 48 hours later showed normal signal characteristics, therefore original changes described may have been artifactual. An EEG showed diffuse slowing consistent with mild encephalopathy. No epileptiform activity was identified. Serology was positive for WNV IgM. Interferon alpha-2b treatment was initiated on admission to Foothills hospital. Over the next few days she gradually regained consciousness and improved. One week following admission, she was awake, alert and interactive, and her strength had returned to an MRC grade of 4+/5 in left leg and 5/5 in all other extremities. Two weeks later she was discharged home ambulating with the aid of a walker, and with mild cognitive slowing and fatigue.

Patient #5

This 71-year-old previously healthy male presented with a two-week history of high fever, mild nuchal headache, diaphoresis and fatigue. With symptom onset, he developed a truncal erythematous maculopapular rash that resolved spontaneously in two days. His other symptoms progressed and he developed disabling ataxia. He described unsteadiness on his feet, disorientation with sudden rotatory movements, and blurry vision. He subsequently developed generalized weakness that lead to hospitalization. On examination he was febrile, slightly hypotensive (90/70), diaphoretic and flushed. Eye movements showed saccadic smooth pursuit, and there was mild right lower facial weakness. All other cranial nerves were normal. Motor examination revealed mild weakness of the right upper and lower extremity with brisk reflexes on the right. A marked left intention tremor was noted, and heel-shin coordination was impaired on the right. The patient was severely ataxic. The remainder of the examination was normal. Brain MRI was normal. Serum was positive for WNV IgM. He was started on treatment with IFN alpha-2b and closely observed. After three days of hospitalization, the patient was subjectively feeling better; his headache and gait had improved. Five days later he was able to walk across the room with only mild residual ataxia. He did not develop any new cranial nerve symptoms; speech, swallowing and vision were normal. His right-sided weakness resolved completely and he was sent home on interferon treatment to complete over two weeks. Two weeks later he developed bilateral leg weakness. An EMG showed evidence compatible with a motor neuronopathy. He gradually improved but remains with mild bilateral leg weakness at four months follow-up.

Patient #6

This 56-year-old man's symptoms began with bitemporal headache, nausea, diarrhea, fatigue, malaise, generalized weakness and fever. He also noticed clumsiness of small co-ordinated movements in his hands. Two days later he experienced twitching in the muscles of his chest, arms and shoulders. He was admitted to hospital for investigation. The following day the twitching resolved but he developed worsening weakness in his left arm. He remained in hospital for five days for observation and supportive treatment, and did not deteriorate or develop new neurological deficits. On discharge his arm strength had slightly improved. He had a persistent mild headache and fatigue. One week later, his fatigue and myalgias continued. His neurological examination revealed severe proximal muscle weakness in the left upper extremity (MRC grading of 2/5 in the deltoid, supraspinatus and infraspinatus), weakness of his left biceps, brachioradialis and forearm supinators (graded 4-/5) and mild weakness in the remaining distal muscles of the left arm and hand (4+/5). The remainder of the neurological examination was normal. His EEG was normal. An MRI of the brain demonstrated nonspecific T2 hyperintensities and cervical spinal cord imaging studies were unremarkable. Serum for WNV IgM was positive, and HI titres increased from 1:40 to 1:640. An EMG demonstrated subacute denervation throughout the left upper extremity; severe in C5 and C6 innervated muscles; moderate in C7; and relatively mild in C8. Sensory studies were normal. Nerve conduction and EMG findings suggested a lower motor neuron (poliomyelitis-like) syndrome. No clinical improvement in left arm weakness was seen one month after onset.

Patient #7

This 50-year-old male presented with a two-week history of flu-like symptoms, followed by red "sheet-like" rash and high fever of 40.4ºC. Two days later, he developed a moderate bifrontal headache, neck pain and stiffness, nausea, vomiting, and blurry vision. His gait was ataxic and he was unable to perform a tandem gait. Serum WNV IgM was positive. Plasma WNV RNA was positive by NASBA. His HI titre rose from less than 1:10 to 1:1280. His symptoms improved over five days and he was discharged home with residual ataxia and blurry vision. However, the following day he awoke with right arm discomfort, and developed sudden facial weakness worse on the right, dysarthria, right palatal weakness, and a bilateral tongue weakness with fasciculations. He was then treated with IFN alpha-2b for progressive delayed onset of predominant brainstem neurological abnormalities. Bulbar findings progressed to bilateral facial paralysis, and worsening dysarthria secondary to palate, tongue and face weakness. Two days post admission, he complained of tingling in the right side of the throat, difficulty swallowing and one night of impaired sleep secondary to breathlessness/gasping with sleep onset. Throughout the course of hospital stay there was no evident extremity weakness. Brainstem symptoms did not progress. Repeat swallowing assessments and spirometry were performed, all of which were normal. Brain MRI showed a few small punctate T2/FLAIR hyperintense foci within the subcortical white matter of both frontal lobes. His symptoms improved over the course of two weeks, the ataxia and tongue weakness completely resolved, palatal elevation had returned to midline and speech improved markedly. This patient was discharged with residual mild improving bilateral facial weakness and fatigue upon completion of interferon treatment. There was no evidence of extremity weakness or new neurological deficits on discharge.

Results

A total of seven patients with WNV neurological syndrome were included in the current study. The time period of viral infection was similar to that of the 1999 New York City outbreak and the 2002 WNV outbreak in Ontario,[9,10] with clinical

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presentations occurring in the later summer and early autumn during the first year of viral activity in these regions. The locations of infection were in keeping with the migratory pattern of WNVacross North America, and the geographical distribution of identified WNV fever in Alberta, where higher numbers of infected individuals have been identified on the southern and eastern borders of the province[11,12] (Table 1).

Three patients were male and four were female. Ages ranged from 24 to 73 years old, with mean age of 52. Six patients were functioning independently with no acute health problems prior to infection. One patient had just completed her fourth chemotherapy (cyclophosamide/adriamycin) cycle for treatment of breast cancer. Six of the seven patients had been participating in outdoor activities prior to infection and four could recall obtaining mosquito bites (Table 2). All patients had flu-like illness and fever as presenting symptoms, and six patients had severe headaches. Fevers ranged from 38.5°C to 40.4°C.

Neurological manifestations of the seven patients with WNV infection are summarized in Table 3. Previous epidemics have identified three classic types of neurological presentation in WNV infection: meningitis/encephalitis, cerebellar symptoms and neuromuscular weakness.[9,10,15-17] In the current case series, two of the seven patients had both encephalitis and neuromuscular weakness, four had cerebellar signs without changes in consciousness, and one patient had focal arm weakness. In three of the four patients with cerebellar findings, a diagnosis of meningitis was also made. The one patient who presented with cerebellar ataxia without meningitis also developed opsoclonus-myoclonus. The four patients with cerebellar ataxia syndromes presented with erythematous or maculopapular rash involving the trunk and extremities that resolved spontaneously. In two cases this rash was pruritic. None of the encephalitic patients reported a history of rash. Two patients had a history of prior cancer (one breast, one chronic lymphocytic leukemia) and were immunocompromised prior to infection. Of these patients, one had severe illness, requiring ventilation support in the ICU, and ultimately died, the only mortality in our series.

Analysis of the CSF was available for five of the seven patients (Table 3). This showed elevation in the white blood cell count in all cases, with ranges from 10.5 to 1139 (x 10^6cells/L). Three cases had a predominantly neutrophilic differential, and in the other two cases the CSF was lymphocytic. Protein was elevated in three of the five cases, the highest value being 1.92gm/L. The CSF cultures were negative in all five cases. West Nile virus RNA was detected by PCR in the CSF of only one patient. Cytological analyses were negative for malignancy in all cases, and all other testing for herpes simplex virus, enterovirus, and other infectious agents that was performed on the different samples were negative.

Electroencephalograms were performed on three of the seven patients and showed diffuse slowing with signs of mild encephalopathy in two cases. Four patients with neuromuscular weakness had nerve conduction and needle EMG findings consistent with lower motor neuron or anterior horn cell lesions. Neuroimaging was performed in all cases. Other than patients 1 (Figure 3) and 4, most brain imaging results were unremarkable. Spinal cord MRI imaging studies in patients 1, 4, 6 and 7 did not demonstrate signal hyperintensities in the anterior horn cell region as described in some of the previous reviews of patients with flaccid paralysis.[15-17] In patient 1 MRI brain imaging demonstrated signal changes in temporal lobes, thalami, substantia nigra, pons and medulla. Patients 4 and 7 showed a relatively nonspecific increase of T2 hyperintensity signal in the subcortical white matter regions bilaterally.

Of the two patients with encephalitis and neuromuscular weakness, one was treated with INF alpha-2b and subsequently recovered. This patient regained consciousness and strength within one week of treatment initiation and was left with mild residual leg weakness, cognitive slowing and logopenia upon discharge. The patient who died had not received treatment due to late presentation to hospital in an already unresponsive state. Two patients with cerebellar signs (one of whom had opsoclonus-myoclonus) improved spontaneously and exhibited only mild residual deficits on discharge. The other two patients with cerebellar symptoms developed brainstem involvement, one coinciding with and one subsequent to, the cerebellar symptoms. These latter two patients 5 and 7 were treated with INF alpha-2b due to ongoing evolution of symptoms. Within one week of treatment these two patients showed marked improvement. Patient 5 developed a delayed poliomyelitis syndrome several weeks after initial symptom improvement.

Discussion

This case review of seven patients in the Calgary region with WNVreports findings similar to those previously reported North American outbreaks [28,29] with some unique features. Multiple neurological symptoms occurred in each patient and the neurological presentation was varied. Classically, WNV is known to cause a combination of meningitis /encephalitis and neuromuscular weakness. In this case series, two patients developed encephalitis and neuromuscular weakness, four patients had cerebellar ataxia plus cranial nerve abnormalities and one patient developed isolated focal extremity weakness. All patients had a flu-like illness prodrome with fever. The incidence of rash (57%) in our study is higher than that (19-26%) reported previously.[9,15] Cerebrospinal fluid analysis was available for five of the seven patients and revealed pleocytosis in all cases, elevated protein in four out of five cases, and normal glucose levels in all cases, findings that are consistent with prior literature.[10,30]

Patient 6 of the current series is a male who presented with focal muscle weakness of one extremity, in a proximal greater than distal distribution. He did not develop meningo-encephalitis but he did complain of subjective fatigue and weakness that persisted at follow-up examination. This would have previously been regarded as an atypical neuromuscular manifestation of WNV and may have guided clinicians toward alternative diagnoses. In our patient, the nerve conduction and EMG findings were consistent with anterior horn cell localization of the lesion. In Leis et al,[29] two similar patients are described, one where flaccid paralysis was confined to a single limb and persisted, and another where transient muscle weakness of the left upper extremity developed and completely resolved within weeks. Recognition of this presentation will prevent misdiagnosis and avoid inappropriate treatment.

It is well-known that weakness is a common manifestation of WNV.[29] In the New York City outbreak of 1999, ten percent of

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patients presented with acute flaccid paralysis and greater than fifty percent of cases involved weakness.[9] It was previously thought that these patients were exhibiting a Guillain-Barré-like syndrome.[31] However recent studies have convincingly identified that WNV can commonly cause a poliomyelitis-like syndrome.[32] Electrodiagnostic studies display features of anterior horn cell or motor axon involvement with reduced or absent compound muscle action potentials in paretic limbs, asymmetric denervation on needle EMG studies and sensory nerve action potentials usually preserved.[15-17] West Nile virus causes inflammation of the spinal cord gray matter during the acute phase of infection leaving peripheral nerves relatively spared. A review of the literature reveals WNV can manifest clinically as a spectrum of weakness including acute flaccid paralysis, reversible muscle weakness and generalized subjective fatigue.[29] Prior reports of patients with WNV associated poliomyelitis described normal MRI studies of the spine and brain.[32] No information on spine imaging is available from the 1999 New York outbreak review. In a recent case series report, three of eight patients showed signal changes involving the parenchyma (conus or cervical cord), cauda equina, or both, usually seen on FLAIR and often enhancing.[15]

Patient 1 was immunocompromised with increased signal change on MRI imaging seen throughout the brainstem and temporal lobes (Figure 2). In the 2002 Ontario case series, MRI of the brain (in 24 patients) showed no acute change except for similar MRI findings to our patient 1 seen in two of their immunocompromised patients.[10] In a recent case series, six of 18 brain imaging studies showed abnormalities either involving cortex or brainstem in isolation or cortex plus underlying subcortical white matter. In this latter series there appeared to be no correlation of imaging findings with immunocompromised status.[15]

The WNVoutbreak in Ontario alerted clinicians to the risk of symptom evolution subsequent to discharge from hospital. Patient 7 initially presented with WNVmeningitis and cerebellar ataxia, partially recovered from these symptoms and subsequently developed bilateral facial palsies, tongue paresis, dysarthria and upper extremity paraethesias. His facial weakness progressed to complete paralysis; however he did not develop any extremity weakness. This presentation suggests a spread of viral activity from the leptomeninges and cerebellum to the brainstem, a preferred site for WNV.[33] An alternative explanation for this clinical presentation might include delayed neuronal dysfunction or death of infected neurons. Needle EMG studies in this patient showed profound denervation of the facial muscles, consistent with a lower motor neuron pattern. This is consistent with the previous reports by Jeha et al[15] identifying the facial nerve as the most commonly affected cranial nerve affected. Patient 5 developed lower motor neuron symptoms several weeks after recovery of other symptoms. These cases illustrate persistence of active infection and evolution of neurological symptoms stressing that regular neurological follow-up is warranted in these patients.

Patient 2 of our series presented with signs so unusual of WNV that this diagnosis was disregarded on admission. This patient presented with opsoclonus-myoclonus in addition to meningitis and cerebellar ataxia following the typical flu-like prodrome. There have been no reports in the literature to date of opsoclonus occurring in WNVinfected patients; and this finding led to the consideration of multiple other diagnoses. Ultimately, serology and CSF analysis of this patient was negative for various infectious etiologies and a diagnosis of WNV was revealed. This case reinforces the fact that WNVinfection of the nervous system can manifest in a variety of signs and symptoms and that clinicians must remain cognisant of atypical presentations when considering the diagnosis. This patient was seen in follow-up two weeks later and had persisting fatigue and anxiety preventing return to work. Generalized weakness and disabling fatigue has also been previously identified as a symptom of WNV infection[29] and, although anxiety is an unusual persisting symptom following the resolution of acute illness, depression has previously been reported in this context.[34]

Persisting symptoms were a universal finding amongst our patients. Although all six patients who had survived infection were markedly improved upon discharge, none had returned to their functional level prior to hospitalization. Few data exist regarding long-term morbidity after hospitalization for WNV infection; those that do suggest that many patients have substantial morbidity. Among patients hospitalized in New York and New Jersey in 2000, more than half did not return to their functional level by discharge and only one third were fully ambulatory.[34] Neurologic symptoms and functional limitations may persist. A one-year follow-up study of the New York outbreak in 1999 revealed that persisting symptoms were present in the majority of patients (fatigue 67%, memory loss 50%, difficulty walking 49%, muscle weakness 44%, and depression 38%).[22,34] These findings encourage public health preventative measures to prevent future infections and therefore decrease the potential morbidity associated with WNV infection. Patient fatalities from WNV neurological syndrome were 12% and 18% in the New York City and Ontario outbreaks respectively.[9,10] In our small series, the mortality rate was 14.5%.

All patients in this study were positive (concordant) in both EIA IgM assays, even in the face of moderate immunosuppression, confirming the clinical utility of the new kits. The PCR assay on CSF had a very low yield for this series, with only two of six patients tested reading weakly positive for viral RNA. This is in contrast to previous reports[35,36] and to the plasma NASBA assay, which performed well in West Nile Fever patients (P. Tilley, in preparation). The EIA assays were not employed for detection of IgM in CSF, as they had not been validated for this purpose to date. Further publications are required to validate the available assays on different specimens.

To date there are no proven treatments for WNV infection. Promising results of ribavirin and INF alpha-2b in laboratory cell-cultures have led to the initiation of a randomized clinical trial of interferon in New York.[20] Interferon alpha-2b has been shown to possess greater therapeutic activity *in vitro* than ribavirin, and has a greater therapeutic ratio in humans,[19] and has been previously used in the treatment of Hepatitis C infection.[27] Furthermore, the side effect profile of this agent is known to be mild and reversible with cessation of treatment.[20] Due to the significant morbidity and risk of mortality associated with WNV infection, we offered this relatively safe treatment to patients who showed little sign of spontaneous improvement with supportive treatment and were deemed to be at significant risk of deterioration.

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Patients 4, 5 and 7 were each given three million units of IFN alpha-2b subcutaneously once per day for a two week duration. All three patients improved following treatment. The marked improvement in the three patients who received INF alpha-2b in the current case series raises preliminary optimism towards this potential treatment. Nonetheless, it remains possible that these patients would have improved spontaneously without the use of interferon and solely with the continuation of supportive treatment. It is expected that the clinical trial currently underway will clarify such issues and determine if this treatment is truly beneficial.

Until the completion of the ongoing randomized trial for the use of IFN alpha-2b in the treatment of WNV encephalitis, the most effective management of WNV today is prevention.

Physicians should warn patients during late summer and early fall to avoid outdoor activity between dusk and dawn, the time when mosquitoes are most abundant. When outdoors, long sleeved shirts and long pants should be worn and mosquito repellent containing DEET should be used.[2,22] Furthermore, physicians must remain vigilant[37] and consider WNV infection as a possible causative agent in patients who present with fever, rash, headache and a variety of different neurological symptoms including weakness, altered level of consciousness and cerebellar signs. All patients with suspected WNV encephalitis should be hospitalized for observation and supportive treatment and other conditions that cause similar presentations must be ruled out.[14] As previously suggested in the literature, the WNVis here in North America to stay.[38] The knowledge gained from the current small case series regarding the variability in presentation and clinical course of WNV neurological syndrome, can help prepare physicians for future outbreaks and this knowledge might minimize the future morbidity and mortality associated with this pathogen.

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