Open Forum Infectious Diseases

MAJOR ARTICLE

The WEST Study: A Retrospective and Multicentric Study on the Impact of Steroid Therapy in West Nile Encephalitis

Marta Colaneri,[1] Raffaella Lissandrin,[1] Matteo Calia,[1] Cecilia Bassoli,[1] Elena Seminari,[1] Alessandro Pavesi,[1] Francesca Rovida,[2] Fausto Baldanti,[2] Alba Muzzi,[3] Guido Chichino,[4] Angelo Regazzetti,[5] Cecilia Grecchi,[5] Angelo Pan,[6] Matteo Lupi,[6] Erica Franceschini,[7] Cristina Mussini,[7,8] and Raffaele Bruno[9]

[1]Division of Infectious Diseases I, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy, [2]Microbiology and Virology Unit, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy, [3]Medical Direction, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy, [4]Division of Infectious Diseases, ASST Ospedale of Alessandria, Alessandria, Italy, [5]Division of Infectious Diseases, Ospedale Delmati di Sant'Angelo Lodigiano, Lodi, Italy, [6]ASST di Cremona, Cremona, Italy, [7]Department of Infectious Diseases, Azienda Ospedaliero-Universitaria "Policlinico of Modena," Modena, Italy, [8]Department of Surgical, Medical, Dental and Morphological Sciences with Interest Transplant, Oncological and Regenerative Medicine, University of Modena and Reggio Emilia, Modena, Italy, and [9]UOC Malattie Infettive, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy

Background. The use of steroid therapy in potentially life-threatening neuroinvasive forms of West Nile infection (WNND) is controversial. The aim of this study is to assess the efficacy of steroid therapy in reducing intrahospital mortality, length of stay, and neurological sequelae at discharge.

Methods. This was a multicenter, retrospective, observational study conducted in 5 hospitals in Northern Italy, headed by the Fondazione IRCSS Policlinico San Matteo (Pavia). We extracted all patient data with WNND diagnoses, comparing patients who received steroid treatment with patients who did not receive steroid treatment between January 2014 and January 2022. Comparisons between the 2 groups were performed using chi-square tests for categorical variables and Mann-Whitney tests for non-normal continuous data, and a generalized linear model for the binomial family was carried out.

Results. Data from 65 WNND patients were extracted. Among these patients, 33 (50.7%) received steroid therapy at any point during their hospitalization. Receiving steroid therapy did not significantly reduce intrahospital mortality (odds ratio [OR], 1.70; 95% CI, 0.3–13.8; $P = .89$) or neurological sequelae at discharge (OR, 0.53; 95% CI, 0.16–1.76; $P = .47$).

Conclusions. Steroid treatment is currently used on a single-case basis in severe WNND. More prospective data are needed to demonstrate a protective effect on mortality and neurological sequelae.

Keywords. West Nile virus; immunocompromised patients; steroid therapy; West Nile virus neuroinvasive disease.

The West Nile virus (WNV) infection is widely distributed in Europe and might result in a broad spectrum of clinical manifestations from asymptomatic disease to potentially life-threatening neuroinvasive forms (WNND) [1].

While WNND is often associated with high mortality rates, especially in older and multipathological patients [2], no specific and effective treatment exists, and supportive care remains the mainstay of treatment [3].

Use of steroids in WNND patients may improve their clinical outcome by reducing host inflammatory response accounting for neurological morbidity. However, the available data are limited and controversial. Several case series have described a clinical improvement after administration of steroids [4, 5], while other authors have failed to conclude the same benefits [3].

Despite a general reluctance to use corticosteroid therapy in infections, it is commonly used in other infections of the CNS, as in bacterial meningitis [6] and herpes simplex virus encephalitis [7].

The aim of this multicenter study was to evaluate the impact of steroid treatment in patients affected by WNND hospitalized in 5 hospitals in Northern Italy. Specifically, we aimed to assess the steroid treatment's effect on mortality rate, length of stay (LOS), and residual neurological sequelae at the time of discharge.

METHODS

Study Design and Setting

The WEST study was a multicenter, observational, retrospective cohort study conducted in 5 hospitals in Northern Italy (IRCCS Fondazione Policlinico San Matteo of Pavia, Ospedale Civile SS Antonio Biagio e Cesare Arrigo of Alessandria, Azienda Socio-Sanitaria Territoriale of Cremona, Ospedale Maggiore of Lodi, and AOU Policlinico of Modena).

This study was a retrospective, multicenter analysis of patients with a confirmed diagnosis of WNND referred to these hospitals from January 2014 to January 2022.

Received 30 November 2022; editorial decision 14 February 2023; accepted 17 February 2023; published online 20 February 2023

Correspondence: Marta Colaneri, MD, Division of Infectious Diseases I, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy (marta.colaneri@gmail.com)

Open Forum Infectious Diseases®

https://doi.org/10.1093/ofid/ofad092

The medical records of all the adult patients with clinical and microbiological diagnoses of WNND were anonymized and abstracted on standardized data collection forms. Demographic and clinical data were retrospectively extracted from both paper and electronic records from the 5 involved hospitals.

Patient Consent

This research project was carried out in accordance with a research plan and according to the current version of the Declaration of Helsinki (https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects).

The sponsor (IRCCS Policlinico San Matteo Foundation, Pavia) ensured that approval from a Competent Ethics Committee (CEC) was sought for the study. All participants signed, before the initiation of treatment and data collection, an informed consent provided by the Fondazione IRCCS Policlinico San Matteo at hospitalization. The WEST study was approved by the IRCCS Policlinico San Matteo Foundation Institutional Review Board (n.prot 0036339/22).

Patient Characteristics

The demographic data included sex and age. Clinical data included symptoms at presentation, comorbidities (history of cancer, heart disease, hypertension, diabetes, chronic kidney disease, lung disease, and obesity), and presence of coinfections. Performed examinations included lumbar puncture (LP) and brain imaging, in the form of computed tomography (CT) scan and magnetic resonance imaging (MRI). Treatment data included the use of antibiotic, antiviral, and steroid treatment.

Steroid therapy was started on the basis of individual clinical judgment in all the included centers. In fact, due to the lack of definite practice guidelines, each physician independently adopted a personal approach according to his own experiences as to whether to administer steroid therapy.

Laboratory Investigation

Serum and cerebrospinal fluid (CSF) samples of patients with potential WNND were tested for the presence of specific immunoglobulin (Ig)M and IgG antibodies (WNV IgM Capture DxSelect and WNV IgG DxSelect by Focus Diagnostics, Cypress, CA, USA). Furthermore, the presence of WNV-specific antibodies was confirmed by a plaque-reduction neutralization test (PRNT). Serum, CSF, and urine samples, collected during the acute phase, were also examined for the presence of WNV RNA with 2 methods: a real-time reverse transcriptase polymerase chain reaction (RT-PCR) targeting a conserved region of WNV lineages 1 and 2 and a pan-*Flavivirus* nested RT-PCR followed by sequencing of amplicons.

Definition of Human WNV Infection

In agreement with the clinical and diagnostic criteria for probable and confirmed WNV case definition, a case was considered probable if the patient met any of the following clinical criteria: suggestive clinical presentation (encephalitis, meningitis, fever ≥38°C) in the presence of WNV-specific IgM or IgG in serum with IgG seroconversion or a 4-fold increase in IgG titer on 2 subsequent samples. A confirmed case was defined as meeting the previous clinical criteria with 1 or more of the following additional criteria: (1) isolation of WNV from blood or cerebral spinal fluid (CSF), (2) detection of WNV-RNA in blood and/or CSF and/or urine, (3) WNV-specific IgM in CSF, and (4) detection of WNV IgM at a high titer and detection of WNV IgG confirmed by neutralization assay.

Outcomes

The primary outcome was to evaluate the impact of the steroid treatment in reducing the intrahospital mortality of hospitalized patients affected by WNND. In-hospital mortality was considered only if related to WNND.

The secondary outcome was to evaluate the effect of steroid treatment in reducing LOS and cumulative incidence of neurological sequelae at the time of discharge.

Statistics

Qualitative variables were described as counts and percentages of each category. Quantitative variables were summarized as mean and SD, or median and interquartile range (IQR), according to the variable distribution. The Shapiro-Wilk test was applied to test the normal distribution of quantitative variables.

Intrahospital mortality and cumulative incidence of neurological sequelae were compared between 2 groups of patients (patients receiving steroid therapy vs patients not receiving steroid therapy) by Pearson chi-square (χ^2) tests. In addition, a generalized linear model for the binomial family was carried out in order to compare in-hospital mortality in the 2 groups of patients, adjusting for demographic and clinical features (age, gender, immunocompromised status, presence of bacterial and viral coinfection, and cells in CSF).

The LOS of the 2 groups of patients (patients receiving steroid treatment vs patients not receiving steroid treatment) was compared by Mann-Whitney test.

The results are reported as odds ratio (OR) and 95% CI. Statistical analyses were conducted using R (version 4.1.2).

RESULTS

Data from 65 patients with WNND were extracted (18 in IRCCS Policlinico San Matteo of Pavia, 7 in Ospedale di Cremona, 6 in Ospedale Civile Santi Antonio e Biagio e

Cesare Arrigo of Alessandria, 7 in Ospedale Maggiore of Lodi, and 27 in Policlinico of Modena).

Thirty-three patients (50.7%) received steroid treatment at any point during their hospitalization as part of WNND management. Dexamethasone was administered in 21 patients (64%), methylprednisolone in 4 patients (12%), and prednisone in 8 patients (24%). The mean dose of administered steroid treatment, referred to in mg of dexamethasone, was 13.6 mg per day.

LP was performed in 56 patients (86%), and CSF appearance was clear with moderate lymphocytic increased cellularity and increased CSF protein in most cases.

Patient characteristics are shown in Table 1.

In total, 10 cases (15.4%) with coinfections were observed. Specifically, 4 (12.5%) were in patients not receiving steroid treatment, and 6 (18.2%) were in patients receiving steroid treatment ($P = .79$). Bloodstream infections (BSIs) represented half of these cases, followed by CNS, respiratory, and urinary tract infections. *Candida albicans* was found as the main responsible pathogen (Table 2).

An MRI was performed on the totality of the WNND patients, but focal MRI lesions were observed in only 10 cases (6.2% in patients receiving steroid treatment and 24.2% in patients not receiving steroid treatment; $P = .10$). These lesions consisted of hyperintense alterations of signal (in long TR sequences) located in temporal, frontal, parietal cortex, and subcortical white matter, para-hippocampus, cerebral peduncles, internal capsule, basal ganglia, thalamus, pons, cerebellar cortex and subcortical white matter, vermis, dentate nuclei, and cerebellar peduncles.

Most patients (50, 77%) received empirical antiviral therapy with acyclovir. In particular, this drug was prescribed to 24 patients (75%) who did not receive steroid therapy and to 26 patients (79%) receiving steroid therapy.

Outcomes

Regarding outcomes, 8 patients (12.3%) died during hospitalization, 4 in each group (patients receiving and not receiving steroid treatment). No difference was observed in cumulative incidence of neurologic sequalae at discharge between the 2 groups. Unexpectedly, LOS was shorter for those patients not receiving steroid treatment (10.8 vs 18.5 days; $P = .0008$).

Similarly, after accounting for potential confounders (age, gender, immunocompromised status, presence of bacterial and viral coinfection, and number of cells in CSF), the multivariate logistic regression model showed that steroid treatment did not have a significant impact on intrahospital mortality (OR, 1.70; 95% CI, 0.3–13.8; $P = .89$) or neurological sequelae at discharge (OR, 0.53; 95% CI, 0.16–1.76; $P = .47$) (Table 3).

Table 1. General and Clinical Characteristics at Time of Clinical Presentation, Diagnosis and Outcomes Data of WNND Patients According to Steroid Treatment

	No Steroid (n = 32)	Steroid Treatment (n = 33)	*P* Value
Demographics			
Age, mean (SD), y	69.7 (16.8)	68.1 (16.7)	.71
Sex, male, No. (%)	24 (75)	24 (73)	1
Comorbidities, No. (%)			
Diabetes	6 (19)	10 (30)	.42
CKD	4 (12)	1 (3)	.33
CHD	15 (46.9)	13 (39)	.72
COPD	3 (9)	3 (9)	1
Hypertension	14 (44)	20 (61)	.26
Neoplasia	2 (6)	5 (15)	.45
Immunocompromised status[a]	1 (3)	5 (15)	.21
Clinical presentation, No. (%)			
Fever	31 (97)	32 (97)	1
Epilepsy	3 (9)	2 (6)	.97
Neck stiffness	9 (28)	13 (39)	.48
GCS <9	5 (16)	6 (18)	1
Headache	10 (31)	13 (39)	.67
Nausea	5 (16)	9 (27)	.40
Skin lesions	3 (9)	3 (9)	1
CSF examination, No. (%)			
LP	25 (78.1)	31 (93.9)	.13
CSF appearance			.06
Clear, colourless	25 (78.1)	18 (54.5)	
Turbid	5 (15.6)	14 (42.4)	
CSF mean cell/cc count (SD)	172.7 (380.5)	202.8 (327.3)	.73
Elevated CSF protein, No. (%)	23 (71.9)	27 (81.8)	.51
Positive virological test for WNV infection diagnosis, No. (%)			
CSF WNV PCR	8 (25)	10 (30)	.84
CSF WNV IgM	17 (53)	17 (51)	1
CSF WNV IgG	8 (25)	10 (30)	1
Urine WNV PCR	20 (62)	20 (61)	1
Urine panflavivirus PCR	11 (48)	10 (67)	1
Serum WNV IgM	28 (87)	26 (79)	1
Serum WNV IgG	21 (66)	21 (64)	.54
Plasma WNV PCR	14 (44)	19 (58)	.38
Outcomes			
Intrahospital mortality, No. (%)	4 (12)	4 (12)	1
LOS, mean (SD), d	10.8 (6.2)	18.5 (11.4)	.0007
Neurologic sequelae at discharge, No. (%)	11 (34)	9 (27)	.75

Abbreviations: CHD, chronic heart disease; CKD, chronic kidney disease; COPD, chronic obstructive pulmonary disease; CSF, cerebrospinal fluid; GCS, Glasgow Coma Scale; IgG, immunoglobulin G; IgM, immunoglobulin M; LOS, length of stay; LP, lumbar puncture; PCR, polymerase chain reaction; WNV, West Nile virus.

[a]For "immunocompromised status," we mean the presence of neutropenia, hematological diseases, any active immunosuppressive therapy, solid organ transplantation, and AIDS-defining conditions.

DISCUSSION

In the present study, we did not find a significant impact of steroid treatment on intrahospital mortality and neurological sequelae at time of discharge of WNND patients from 5 different hospitals in Northern Italy. The presence of any coinfection was associated with an increased risk of death, and immunocompromised status increased the risk of neurological sequalae at discharge.

Finally, we found that LOS was significantly longer in those patients who were treated with steroids.

WNND include a mixed pattern of diseases such as meningitis, encephalitis, flaccid paralysis, and other less common neurologic manifestations, which have an overall mortality rate of ~10% [8]. Age, male gender, and immunocompromised status are well-known risk factors for increased risk of death or neurological sequelae at discharge [9, 10]. Our study basically confirms the mortality trend, and it supports the association between immunocompromised status and neurological sequelae but not with intrahospital mortality.

In Italy, WNV is endemic, and WNND cases are clustered in the north, particularly in Po River Valley [11, 12], where the hospitals included in our study are located. Hence, a national WNV surveillance plan was recently implemented, with the aim to integrate veterinary, entomological, and epidemiological data to monitor WN spread in both vectors and humans.

Hence, considering that WNND is a life-threatening disease, apart from such epidemiological approaches, we believe it worthwhile to provide real-life reporting of clinical management of this condition, which might be useful for clinicians' everyday practice.

Following this line of thought, as the actual treatment of WNND primarily consists of supportive care, with some agents that may be of potential benefit such as steroids [13, 5] and intravenous immunoglobulin [14], understanding what might truly be helpful in clinical practice is crucial.

Against the use of steroids in WNND, some clinicians might point to the steroid's immunosuppressive effect, which might promote WN viremia and worsen patients' outcomes. On the other hand, others might mention that WNV infection notoriously causes a significant upregulation of several important proinflammatory molecules, which are associated with severity of the WNV infection itself [15, 16], and therefore might potentially benefit from steroid administration.

Moreover, this uncertainty is reinforced by the conflicting results of some case reports and small studies on the topic [4, 17].

In our study, which, to the best of our knowledge, is the largest specifically addressing the clinical impact of steroids in WNND, half of the clinicians chose to start steroid therapy solely relying on their personal clinical experience.

The 2 groups of patients (receiving and not receiving steroid treatment) were homogenous regarding age, comorbidities, and clinical presentation. It must be underlined that within the group of patients treated with steroids there were more immunocompromised patients, which might lead us to reflect on the physicians' propensity to start steroids in the more fragile patients.

We found that LOS was longer in steroid-treated patients than the non-steroid-treated group. This intriguing result let us speculate on the potential role of prolonged hospital stay in the more complex patients rather than the steroid treatment itself.

Specifically, we believe that the more clinically challenging immunocompromised patients usually need more time to be discharged, especially when they require transfer to other rehabilitation facilities.

Furthermore, CSF had a more turbid appearance in patients who received steroid therapy, which might prompt us to consider the physicians' propensity to start steroids when the

Table 2. Other Microbiological Isolations in WNND Patients According to Steroid Treatment

	No Steroid Group (n = 32)	Steroid Group (n = 33)	Total (n = 65)
Other microbiological isolation, No. (%)	4 (12.5)	6 (18.2)	10 (15.4)
Sample and etiology, No. (%)			
Blood	3 (9.4)	3 (9.1)	6 (9.2)
Candida albicans	1 (3.1)	1 (3.0)	2 (3.1)
EBV	1 (3.1)	0 (0.0)	1 (1.5)
Enterococcus faecium	0 (0.0)	1 (3.0)	1 (1.5)
Acinetobacter baumannii	0 (0.0)	1 (3.0)	1 (1.5)
Strongyloides stercoralis	1 (3.1)	0 (0.0)	1 (1.5)
CSF	0 (0.0)	2 (6.1)	2 (3.1)
EBV	0 (0.0)	2 (6.1)	2 (3.1)
Urine	1 (3.1)	1 (3.0)	2 (3.1)
EBV	1 (3.1)	0 (0.0)	1 (1.5)
Enterococcus faecium	0 (0.0)	1 (3.0)	1 (1.5)

Abbreviations: CSF, cerebrospinal fluid; EBV, Epstein barr virus; HSV-1, herpes virus 1.

Table 3. Multivariate Logistic Regression for Intrahospital Mortality and Neurological Sequelae at Discharge

	Intrahospital Mortality			Neurological Sequalae at Discharge		
	OR	95% CI	*P* Value	OR	95% CI	*P* value
Steroid treatment	1.70	(0.3–13.8)	.89	0.53	(0.16–1.76)	.47
Age	1.51	(1.07–3.11)	.08	1.03	(0.99–1.09)	.09
Sex	6.66	(0.02–12.56)	.94	1.19	(0.27–4.85)	.80
Immunocompromised status	7.47	(0.02–994.41)	.98	17.2	(2.18–233.16)	.01
Presence of coinfections	19.5	(0.18–248.99)	.025	0.13	(0.01–0.85)	.06
CSF cell count	0.99	(0.89–1.00)	.28	1.00	(0.99–1.00)	.10

Abbreviations: CSF, cerebrospinal fluid; OR, odds ratio.

etiology of the neurological infection is still unclear. This is based on the existing broader consensus on the efficacy of steroid treatment in other CNS infections, including bacterial meningitis [18].

The limitations of our study are related to its retrospective nature, which does not allow a long-term follow-up of the examined patients, and the observational study design, which does not allow randomization of the therapeutic choice of steroid treatment. Finally, we did not consider transfer to the ICU, which can also delay the patient's discharge home, thus potentially contributing to the LOS difference found between the 2 study groups.

Nevertheless, we think we can draw important conclusions from these data. The lack of a beneficial effect in terms of both intrahospital mortality and neurological sequelae of steroid therapy should certainly not be ignored, as it is based on real-life clinical data. Instead, it is necessary to build on this for future RCTs that are likely to provide a more accurate answer to this important question, which is entirely in the hands of the individual physician's experience to date.

Acknowledgments

Financial support. No funding was provided.

Potential conflicts of interest. All authors: no reported conflicts.

Ethical approval. The study protocol was approved by the Sponsor center (IRCCS Policlinico San Matteo, Pavia, Italy) ethical committee.

References

1. Bauerfeind R, Von Graevenitz A, Kimmig P, et al. Zoonoses: infectious diseases transmissible from animals to humans, fourth edition. Clin Infect Dis **2016;** 63: 148–9.
2. Gray TJ, Webb CE. A review of the epidemiological and clinical aspects of West Nile virus. Int J Gen Med **2014;** 7:193–203.
3. Bode AV, Sejvar JJ, Pape WJ, Campbell GL, Marfin AA. West Nile virus disease: a descriptive study of 228 patients hospitalized in a 4-county region of Colorado in 2003. Clin Infect Dis **2006;** 42:1234–40.
4. Leis AA, Sinclair DJ. Lazarus effect of high dose corticosteroids in a patient with West Nile virus encephalitis: a coincidence or a clue? Front Med (Lausanne) **2019;** 6:81.
5. Zaltzman R, Klein C, Gordon CR. Opsoclonus myoclonus ataxia associated with West Nile virus infection: a dramatic presentation with benign prognosis? J Neurol Sci **2017;** 376:38–41.
6. Brouwer MC, Mcintyre P, Prasad K, van de Beek D. Corticosteroids for acute bacterial meningitis. Cochrane Database Syst Rev **2015;** 2015:CD004405.
7. Ramos-Estebanez C, Lizarraga KJ, Merenda A. A systematic review on the role of adjunctive corticosteroids in herpes simplex virus encephalitis: is timing critical for safety and efficacy? Antivir Ther (Lond) **2014;** 19:133–9.
8. Petersen LR, Brault AC, Nasci RS. West Nile virus: review of the literature. JAMA **2013;** 310:308–15.
9. Patel H, Sander B, Nelder MP. Long-term sequelae of West Nile virus-related illness: a systematic review. Lancet Infect Dis **2015;** 15:951–9.
10. Visentin A, Nasillo V, Marchetti M, et al. Clinical characteristics and outcome of West Nile virus infection in patients with lymphoid neoplasms: an Italian multicentre study. HemaSphere **2020;** 4:e395.
11. Riccò M, Peruzzi S, Balzarini F. Epidemiology of West Nile virus infections in humans, Italy, 2012–2020: a summary of available evidences. Trop Med Infect Dis **2021;** 6:61.
12. Rovida F, Sarasini A, Campanini G, et al. West Nile virus outbreak in the Lombardy region, Northern Italy, summer 2013. Vector Borne Zoonotic Dis **2015;** 15:278–83.
13. Pyrgos V, Younus F. High-dose steroids in the management of acute flaccid paralysis due to West Nile virus infection. Scand J Infect Dis **2004;** 36:509–12.
14. Gnann JW, Agrawal A, Hart J, et al. Lack of efficacy of high-titered immunoglobulin in patients with West Nile virus central nervous system disease. Emerging Infect Dis **2019;** 25:2064–73.
15. Fraisier C, Papa A, Almeras L. High-mobility group box-1, promising serological biomarker for the distinction of human WNV disease severity. Virus Res **2015;** 195:9–12.
16. Kuwar RB, Stokic DS, Leis AA, et al. Does astroglial protein S100B contribute to West Nile neuro-invasive syndrome? J Neurol Sci **2015;** 358:243–52.
17. Murray KO, Baraniuk S, Resnick M, et al. Clinical investigation of hospitalized human cases of West Nile virus infection in Houston, Texas, 2002–2004. Vector Borne Zoonotic Dis **2008;** 8:167–74.
18. Van De Beek D, De Gans J, McIntyre P, Prasad K. Steroids in adults with acute bacterial meningitis: a systematic review. Lancet Infect Dis **2004;** 4:139–43.