Journal of Microbiology and Infectious Diseases / JMID
2012; 2 (4): 155-159
doi: 10.5799/ahinjs.02.2012.04.0063

ORIGINAL ARTICLE

Evaluation of clinical and laboratory predictors of fatality in patients with Hantavirus infection

Uğur Kostakoğlu[1], Gürdal Yılmaz[2], Serkan Volkan[3], Sevinç Kant Sökel[4], Selçuk Kaya[2], İftihar Köksal[2]

[1] *Kanuni Education and Research Hospital, Department of Infectious Diseases and Clinical Microbiology, Trabzon, Turkey*
[2] *Faculty of Medicine, Karadeniz Technical Univ. Department of Infectious Diseases and Clinical Microbiology, Trabzon, Turkey*
[3] *Aydın State Hospital, Department of Infectious Diseases and Clinical Microbiology, Aydin, Turkey*
[4] *Burdur Health Directorate, Director of Communicable Diseases Unit, Burdur, Turkey*

ABSTRACT

Objectives: To determine the clinical and laboratory predictors of fatality among patients with Hantavirus infection.

Materials and methods: A retrospective study was conducted on the patients with Hantavirus infection between April 2009 and October 2011 at the Black Sea and the Mediterranean regions in Turkey. Demographic, clinical and laboratory findings of fatal cases and non-fatal cases at the admission were compared.

Results: Twenty-two patients with confirmed Hantavirus infection were evaluated. Five patients died (22.7%). The cause of death was massive bleeding. The rate of hemorrhage was significantly higher in the fatal cases than non-fatal cases ($p<0.001$). Massive gastrointestinal hemorrhage was seen in four of these patients, cerebral hemorrhage in two and both gastrointestinal and pulmonary hemorrhage in one. Disseminated intravascular coagulation (DIC) was present in four of the fatal cases and a remarkable cause of the bleeding ($p=0.02$). White blood cell count (WBC) ($p=0.002$), creatine phosphokinase (CPK) ($p=0.011$), blood-urea-nitrogen (BUN) ($p=0.014$), C reactive protein (CRP) ($p=0.005$) and D-dimer levels ($p=0.001$), prothrombin time (PT) ($p=0.023$), activated partial thromboplastin time (aPTT) ($p=0.001$) and international normalized ratio (INR) ($p=0.021$) were significantly higher, and platelet counts ($p=0.038$) significantly lower in the fatal cases. Optimum diagnostic cut-off points for specific laboratory parameters which may be predictive of fatality were; WBC=16,000 µL-1, PLT=30000 µL-1, PT=19.7 s, aPTT=36 s, INR=1.2, D-dimer=9.3 µg/mL, CPK=600 U/L, BUN=47 mg/dL and CRP=13.4 mg/dL.

Conclusions: Physicians should be aware of the high fatality risk for patients with Hantavirus infection with hemorrhage, elevated WBC, CPK, BUN, CRP, PT, aPTT, INR and D-dimer and reduced platelet counts. *J Microbiol Infect Dis 2012; 2(4): 155-159*

Key words: Hantavirus; hemorrhagic fever with renal syndrome; predictors of fatality; predictive factors

Hantavirüs enfeksiyonu olan hastalarda ölüm için klinik ve laboratuvar öngörü faktörlerinin değerlendirilmesi

ÖZET

Amaç: Hantavirüs enfeksiyonu olan hastalar arasında fatalitenin klinik ve laboratuvar belirteçlerini belirlemek.

Gereç ve yöntem: Bu retrospektif çalışma, Türkiye'de Karadeniz ve Akdeniz bölgesinde Nisan 2009 ve Ekim 2011 tarihleri arasında hantavirüs enfeksiyonu olan hasta üzerinde yapıldı. Ölümcül ve ölümcül olmayan vakaların hastaneye kabulde kaydedilen demografik, klinik ve laboratuar bulguları karşılaştırıldı.

Bulgular: Hantavirüs enfeksiyonu teyit edilen yirmi iki hasta incelendi. Beş hasta (% 22,7) öldü. Ölümlerin nedeni masif kanama idi. Ölümcül olmayan vakalarla karşılaştırıldığında ölenlerde kanama oranı yüksekti ($p<0,001$). Bu vakaların dördünde masif gastrointestinal kanama, ikisinde serebral kanama ve birinde hem gastrointestinal hem de pulmoner kanama görüldü. En önemli kanama nedeni dissemine intravasküler koagülasyon (DİK) idi. DİK ölen vakaların dördünde vardı ve mortalite ile ilişkili idi ($p=0,02$). Ölen vakalarda WBC ($p=0,002$), CPK ($p=0,011$), BUN ($p=0,014$), CRP ($p=0,005$)ve D-dimer düzeyleri ($p=0,001$), PT ($p=0,023$), aPTT ($p=0,001$) and INR ($p=0,021$) yüksek, trombosit sayısı ($p=0,038$) düşük idi. Ölüm öngörüsü için laboratuvar parametrelerinin tanısal kesme noktaları WBC=16000 µL-1, PLT=30000 µL-1, PT=19.7 s, aPTT=36 s, INR=1.2, D-dimer=9.3 µg/mL, CPK=600 U/L, BUN=47 mg/dL ve CRP:13,4 mg/dL idi.

Sonuç: Biz, doktorların kanaması olan, artmış WBC, CPK, BUN, CRP, PT, aPTT, INR, D-dimer ve azalmış trombosit sayısı olan hantavirüs enfeksiyonu hastalarında ölüm riskinin yüksek olduğunun farkında olunması gerektiğini düşünüyoruz.

Anahtar kelimeler: Hantavirus; renal sendromlu kanamalı ateş; ölüm göstergeleri; öngörü faktörleri

Correspondence: Uğur Kostakoğlu, Kanuni Education and Research Hospital,
Department of Infectious Diseases and Clinical Microbiology, Trabzon, Turkey Email: ukostakoglu@gmail.com
Received: 10 July, 2012 Accepted: 05 November, 2012

INTRODUCTION

Hantaviruses, a member of the family Bunyaviridae, can lead to two different types of infection in humans; hemorrhagic fever with renal syndrome (HFRS) and hantavirus pulmonary syndrome (HPS).[1] Some 150,000 cases a year are hospitalized globally and treated with a diagnosis of HFRS.[1,2] HFRS is the most common type of Hantavirus infection in Europe and Asia; the most common virus types are Dobrava, Puumala, Hantaan and Seoul.[1,3,4] The HPS infection type, widely seen on the American continent, caused by the virus types Sin Nombre, Andes, Laguna Negra and New York.[5] While these infections may follow mild clinical courses, they are also potentially fatal diseases capable of leading to a severe clinical course accompanied by hemorrhage.[1,3] While HPS has a 35%-50% mortality rate, that in HFRS is 0.1%-15%.1 The ability to determine the clinical course and fatality risk of the disease beforehand may be life-saving by ensuring that the supportive therapy administered to the patient is appropriate.

The aim of this study was to use clinical and laboratory results on admission to determine the clinical and laboratory predictors of fatality among patients with Hantavirus infection.

MATERIALS AND METHODS

This retrospective study was conducted on patients with Hantavirus infection hospitalized between April 2009 and October 2011 at the Burdur State Hospital, Trabzon Kanuni Education and Research Hospital and Karadeniz Technical University Medical Faculty in Turkey. Demographic characteristics, clinical findings and laboratory tests on admission of all patients with Hantavirus infection were investigated. We compared clinical and laboratory findings of the fatal cases with non-fatal ones. Patients whose Hantavirus infection diagnoses were confirmed in the Virology Laboratory of the Refik Saydam National Hygiene Center of the Turkish Ministry of Health were included. Serum specimens were investigated for Hantavirus using the immunofluorescent assay (IFA) technique in a 1:100 serum dilution with Hantavirus mosaic-1 (Euroimmun, Germany) kits for scanning purposes in line with the manufacturer's instructions. Specimens testing positive were confirmed using the immunoblot test. For that purpose, Hanta Profile 1 EUROLINE (Euroimmun, Germany) tests were employed in line with the manufacturer's recommendations.

Patients in whom infectious (Crimean-Congo hemorrhagic fever, hepatitis A, B, and C viruses, herpes viruses, HIV, malaria, brucella, leptospira) and noninfectious diseases (hematologic diseases) were excluded by routine tests. Demographic characteristics, occupation, incubation time, symptoms, duration between onset of symptoms and admission, clinical findings, and laboratory tests (white blood cell count [WBC], platelet count [PLT], hemoglobin [Hb], aspartate aminotransferase [AST], alanine aminotransferase [ALT], total bilirubin, lactate dehydrogenase [LDH], creatine phosphokinase [CPK], blood-urea-nitrogen [BUN] and plasma creatinine [Cr], prothrombin time [PT], activated partial thromboplastin time [aPTT], international normalized ratio [INR]) and (D-dimer) of all patients were recorded on forms on admission.

Statistical Analysis: Descriptive statistics were performed for all the studied variables recorded on admission. The data obtained in measurements were analyzed with the Mann Whitney-U test. Data obtained by measurements were given as mean ± standard deviation. Data obtained by counting were given as numbers (%); analyses were performed using the Chi-square test. The area beneath the receiver operating characteristics (ROC) curves was used to calculate the discriminative power of the biochemical marker to determine the predictive criteria for fatality among patients with Hantavirus infection. Sensitivity, specificity, negative predictive value (NPV) and positive predictive value (PPV) were calculated according to ROC curves for each biochemical marker. Statistical significance was set at $p<0.05$.

RESULTS

Twenty-two patients with confirmed HFRS Hantavirus (Dobrava subtype) were investigated. Nine (40.9%) were female and 13 (59.1%) male. Five of these patients died (22.7%). Ages ranged between 11 and 83, with a mean age of 60.4±9.9 in fatal cases and 45.3±17.0 in non-fatal (P=0.058). Blood and blood products transfusion was administered to seven (31.8%) patients with massive hemorrhage. The cause of death was massive bleeding. Massive gastrointestinal hemorrhage was seen in four of these patients, cerebral hemorrhage in two and both gastrointestinal and pul-

monary hemorrhage in one. The rate of hemorrhage was higher in fatal cases compared with non-fatal cases (p<0.001). Disseminated intravascular coagulation (DIC) developed in 31.8% of patients. DIC was the most important cause of the bleeding. DIC was present in four of the fatal cases and was correlated with mortality (P=0.02). Hemodialysis was administered to nine (40.9%) patients. Administration of hemodialysis was not statistically difference between the groups (P=0.135). WBC (p=0.002), CPK (p=0.011), BUN (p=0.014), CRP (p=0.005) and D-dimer levels (p=0.001), PT (p=0.023), aPTT (p=0.001) and INR (p=0.021) were higher, and platelet counts (p=0.038) lower in the fatal cases.

At evaluation of laboratory findings recorded on admission, using the ROC curve method, which display a statistically significant difference at univariate analysis, the optimum diagnostic cut-off points for specific laboratory parameters of the fatal group were; WBC=16,000 µL-1, PLT=30,000 µL-1, PT=19.7 s, aPTT=36 s, INR=1.2, D-dimer=9.3 µg/mL, CPK=600 U/L, BUN=47 mg/dL and CRP=13.4 mg/dL. The specified cut-off points and the sensitivity, specificity, PPV, NPV, and the area underneath the ROC curve (AUC) for those cut-off points are shown in Table 2.

Table 1. Demographic, clinical and laboratory characteristics of patients with Hantavirus infection

Characteristics	Fatal cases n=5	Non-fatal cases n=17	P value
Age (years)	60.4±9.9	45.3±17.0	0.055
Sex (male/female)	3/2	10/7	0.684
Duration of complaints until hospitalization, (days)	3.8±2.9	5.2±3.3	0.543
Fever	4 (80%)	13 (76.5%)	0.687
Myalgia	5 (100%)	12 (70.6%)	0.235
Headache	4 (80%)	11 (64.7%)	0.477
Exhaustion	5 (100%)	13 (76.5%)	0.325
Nausea	2 (40%)	12 (70.6%)	0.232
Vomiting	2 (40%)	10 (58.8%)	0.406
Hemorrhage	5 (100%)	2 (11.8%)	<0.001
Laboratory findings			
WBC (μL^{-1})	22600±4833	10200±3716	0.002
Hb (g/dL)	11.9±2.7	13.6±2.2	0.256
PLT (μL^{-1})	47600±37031	87882±34106	0.038
PT (s)	21.3±5.7	14.5±2.4	0.023
aPTT (s)	57.7±13.3	36.7±3.0	0.001
INR	1.48±0.39	1.05±0.09	0.021
D-Dimer (µg/mL)	28.8±8.6	4.4±4.2	0.001
AST (U/L)	156±172	176±365	0.183
ALT (U/L)	67±58	178±391	0.583
LDH (U/L)	872±625	494±403	0.108
CPK (U/L)	2827±4672	378±846	0.011
BUN (mg/dL)	72.6±27.4	31.4±23.7	0.014
Cr (mg/dL)	4.0±2.6	2.5±2.4	0.136
CRP (mg/dL)	28.2±9.2	11.4±7.6	0.005
Total Bilirubin (mg/dL)	1.0±0.2	0.9±1.4	0.059

Normal ranges: White Blood Cell count [WBC]= 4000-10000 µL-1, Platelet count [PLT]=140,000-500,000 µL-1, Hemoglobin [Hb]=12-18 g/dL, Aspartate aminotransferase [AST]=5-40 U/L, Alanine aminotransferase [ALT]=0-40U/L, Total Bilirubin=0-1.2 mg/dL, Lactate dehydrogenase [LDH]=123-243 U/L, Creatine phosphokinase [CPK]=25-200 U/L, Blood-urea-nitrogen [BUN] and plasma Creatinine [Cr], Prothrombin time [PT]=11.5-15.5 s, activated Partial Thromboplastin time [aPTT]=20-34 s, International normalized ratio [INR]=0.8-1.2, D-dimer=0-0.5 µg/mL.

Table 2. Cut-off points of laboratory findings in patients with Hantavirus infection which may be predictive of fatality, and the AUC, sensitivity rates, specificity rates, PPV, and NPV of those cut-off points

Laboratory findings	Cut-off point	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	AUC
WBC	16000	80	100	100	94.4	0.976
PLT	30000	60	88.2	60	88.2	0.812
PT	19.7	60	100	100	88.2	0.847
aPTT	36	100	93.3	83.3	100	0.986
INR	1.2	80	100	100	93.7	0.853
D-dimer	9.3	100	93.3	83.3	100	0.986
CPK	600	60	88.2	60	88.2	0.859
BUN	47	80	75	50	92.3	0.862
CRP	13.4	100	87.5	71.4	100	0.937

Normal ranges: White Blood Cell count [WBC]= 4000-10000 µL-1, Platelet count [PLT]=140,000-500,000 µL-1, Hemoglobin [Hb]=12-18 g/dL, Aspartate aminotransferase [AST]=5-40 U/L, Alanine aminotransferase [ALT]=0-40U/L, Total Bilirubin=0-1.2 mg/dL, Lactate dehydrogenase [LDH]=123-243 U/L, Creatine phosphokinase [CPK]=25-200 U/L, Blood-urea-nitrogen [BUN] and plasma Creatinine [Cr], Prothrombin time [PT]=11.5-15.5 s, activated Partial Thromboplastin time [aPTT]=20-34 s, International normalized ratio [INR]=0.8-1.2, D-dimer=0-0.5 µg/mL.

DISCUSSION

The prevalence of Hantavirus infection is increasing every day and it is classified among endemic infections in many countries of the world. Studies in various countries have reported a seroprevalence of Hantavirus infections of 0%-33.3%.[1,6-8] These infections are seen in forested regions with plentiful rainfall containing large rodent populations.[4] Following an incubation period HFRS clinical manifestations, although not always typically distinct, consecutively evolve through febrile (prodrome), hypotensive, oliguric, polyuric and convalescent phases; laboratory and imaging findings have been correlated with disease phases.9 Patients may present with mild clinical findings or with potentially fatal severe ones.[1] Patients may frequently exhibit symptoms such as fever, lethargy, muscle pain, nausea and vomiting. Hemorrhage, hypotension, oliguria-anuria and proteinuria may be seen in severe cases.[1,10] Supportive therapy is the most important component of Hantavirus infection management and may involve intensive care.[1,11] Patients must be monitored and supported in terms of vital findings and hemodynamic values.[1,7,10,11] When necessary, hemodialysis and blood-blood product transfusion must be performed. Hemorrhage was seen in 31.8% of our patients; blood-blood products transfusion was performed, and hemodialysis was administered to 40.9% of cases. In Maftei study, transfusion was required in five (83%) out of six patients, and this was correlated with severity of the disease.[11] Hukić et al. reported that Acute haemodialysis was needed in 28% of the patients infected with Dobrava virus.[12]

A mortality rate of 0.1%-15% has been reported in HFRS patients.[1-3,6,11,13] However, 22.7% of the patients in our study died. Our high mortality rate may have been due to our low total patient numbers, the presence of massive hemorrhage and DIC in the fatal cases, and these patients being elderly and having underlying diseases. It has been reported that DIC may develop in 27%-28% of HFRS cases and that it is correlated with mortality.[13,14] DIC developed in 31.8% of our patients and was also correlated with mortality. This high mortality rate shows that the disease is a potentially fatal one and that supportive treatment must be very well administered. Vital parameters and hemodynamic values should be closely monitored in such patients. Adequate support must be provided, and if necessary they should be transferred to the intensive care unit.[1] The physician should also monitor hematological parameters, and replacement therapy for platelet, erythrocyte and coagulation factors must be performed if appropriate.

Elevated serum WBC, CRP, CPK, BUN and Cr are some of the findings that may be observed in Hantavirus infections.[11,13] Our aim was to determine the predictive criteria for fatality in HFRS and thus fill a significant gap in the relevant lit-

erature. Presence of hemorrhage, a low platelet count, prolonged PT and aPTT, and high INR, D-Dimer, BUN, CPK, and CRP were determined as predictors for fatality using univariate analysis.

Renal failure is a significant finding in HFRS. Previous studies have reported that creatinine level is a marker for the clinical severity of the disease.[13-15] While creatinine level was not correlated with mortality, serum BUN levels were high in our fatal cases. We established that BUN level at the determined cut-off point can predict mortality at 80% sensitivity and 75% specificity and 92.3% NPV.

A rise in serum WBC and CRP is correlated with the inflammatory process triggered by cytokines developing during the disease.[1] Elevated CPK is probably related to this inflammatory process and disease-associated rhabdomyolysis. It is noteworthy that serum WBC, CRP and CPK levels during hospitalization in our fatal cases were significantly higher compared to those of the patients discharged in a healthy condition. This elevation may be interpreted as a marker of intense inflammation and severe disease. At the determined cut-off points CRP is capable of predicting mortality with 100% sensitivity and 87.5% specificity, and CPK at 60% sensitivity and 88.2% specificity.

In conclusion, a number of biochemical and hematological parameters that can easily be measured as patients with suspected Hantavirus infection undergo routine examination are useful and sensitive predictors. These can be of particular use in identifying potentially fatal Hantavirus infection. They can also represent a useful addition to supporting treatment and, if required, the appropriate intensive care services. Prior prediction will enable physician to make the most effective selection and use of the available options.

Acknowledgments

This study is presented as poster presentations 4.EACID Congress

Funding: We have not financial support

Competing interests: We have not conflict of interest

Ethical approval: The study had been approved with 33-2011 reference number by Local Ethical Committee.

REFERENCES

1. Jonsson CB, Figueiredo LT, Vapalahti O. A global perspective on hantavirus ecology, epidemiology, and disease. Clin Microbiol Rev 2010; 23:412-441.
2. Song, G. Epidemiological progresses of hemorrhagic fever with renal syndrome in China. Chin Med J (Engl.) 1999; 112:472-477.
3. Heyman P, Vaheri A, Lundkvist A, Avsic-Zupanc T. Hantavirus infections in Europe: from virus carriers to a major public-health problem. Expert Rev Anti Infect Ther 2009; 7:205-217.
4. Heyman P, Vaheri A; ENIVD Members. Situation of hantavirus infections and haemorrhagic fever with renal syndrome in European countries as of December 2006. Euro Surveill 2008; 13 (28). pii: 18925.
5. Simpson SQ, Spikes L, Patel S, Faruqi I. Hantavirus pulmonary syndrome. Infect Dis Clin North Am 2010; 24:159-173.
6. Muranyi W, Bahr U, Zeier M, van der Woude FJ. Hantavirus infection. J Am Soc Nephrol 2005; 16:3669-3679.
7. Vapalahti O, Mustonen J, Lundkvist A, Henttonen H, Plyusnin A, Vaheri A. Hantavirus infections in Europe. Lancet Infect Dis 2003;3:653-661.
8. Ertek M, Buzgan T; Refik Saydam National Public Health Agency. An outbreak caused by Hantavirus in the Black Sea Region of Turkey, January-May 2009. Euro Surveill 2009; 14(20).
9. Sargianou M, Watson DC, Chra P, et al. Hantavirus infections for the clinician: From case presentation to diagnosis and treatment. Crit Rev Microbiol 2012; 38:317-329.
10. Kaya S, Yılmaz G, Erensoy S, Yağçı ÇD, Uyar Y, Köksal I. Hantavirus infection: two case reports from a province in the Eastern Black Sea Region, Turkey. Mikrobiyol Bul 2010;44:479-487.
11. Maftei ID, Segall L, Gatej RP, Ceianu C, Covic A. Hantavirus infection-Hemorrhagic fever with renal syndrome: the first case series reported in Romania and review of the literature. Int Urol Nephrol 2012; 44:1185-1191.
12. Hukić M, Tulumović D, Calkić L. The renal failure and capillary leak during the acute stage of (Dobrava) DOB and PUU (Puumala) infection. Med Arh 2005; 59:227-230.
13. Takeuchi T, Yamamoto T, Itoh M, Tsukada K, Yasue N, Lee HW. Clinical studies on hemorrhagic fever with renal syndrome found in Nagoya City University Medical School. Kidney Int Suppl 1991;35:84-87.
14. Sundberg E, Hultdin J, Nilsson S, Ahlm C. Evidence of disseminated intravascular coagulation in a hemorrhagic fever with renal syndrome-scoring models and severe illness. PLoS One 2011; 6:e21134. Epub 2011 Jun 23.
15. Rasche FM, Uhel B, Krüger DH, et al. Thrombocytopenia and acute renal failure in Puumala hantavirus infections. Emerging Infectious Diseases 2004; 10: 1420-1425.