MAJOR ARTICLE

Risk Parameters of Fulminant Acute Respiratory Distress Syndrome and Avian Influenza (H5N1) Infection in Vietnamese Children

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A clinical picture of patients with acute respiratory distress syndrome (ARDS) induced by highly pathogenic avian influenza A (H5N1) has been reported. We reviewed 37 sets of clinical data for pediatric patients with ARDS at the National Hospital of Pediatrics (Hanoi, Vietnam); 12 patients with H5N1-positive and 25 with H5N1-negative ARDS were enrolled. The H5N1-negative patients had a clinical picture and mortality rate similar to that for the pediatric ARDS patients. However, the H5N1-positive patients had ARDS with normal ventilation capacity at the time of hospital admission, then rapidly proceeded to severe respiratory failure. The survival probability and days until final outcome in groups of H5N1-positive ($n = 12$) vs. H5N1-negative ($n = 25$) patients were 17% versus 52% and 12.3 ± 5.7 days (median, 11 days) versus 21.5 ± 13.8 days (median, 22 days), respectively. Our observations clarified the clinical picture of H5N1-induced fulminant ARDS and also confirmed that relatively older age (~6 years of age), high fever at onset, and leukopenia and/or thrombocytopenia at the time of hospital admission are risk parameters for H5N1-induced fulminant ARDS.

Highly pathogenic avian influenza A (H5N1) came to the attention of the international scientific community for the first time in 1997 [1, 2]. The current global spread of human infection by this subtype started in 2003 in Hong Kong [2, 3], during the global outbreak of severe acute respiratory syndrome [4, 5]. Vietnam reported the first human case of H5N1 infection in January 2004 [6] and a suspected human-to-human transmission family cluster in the following months [7]. Since then, many clinical case reports have been reported from several countries, such as Thailand, Indonesia, and Vietnam [8–14]. However, it is still difficult to detect most infection at first examination without a clear history of patient contact with sick poultry.

The fatality rate associated with pediatric acute respiratory distress syndrome (ARDS) has decreased during recent decades because of advances in medical treatment, especially respiratory management as a lung-protective therapy [15]. However, the majority of patients with H5N1 subtype influenza virus infection experienced or presented ARDS during their clinical courses, often followed by a serious outcome. The histopathology of these cases demonstrated diffuse alveolar damage in the lung, which also suggests ARDS as a clinical condition of the respiratory system [16–18]. Because of the significant possibility that H5N1 subtype influenza will the source of the next pandemic influenza strain [19, 20], the pathophysiology of the clinical course of H5N1

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Table 1. Clinical Features of H5N1-Positive and H5N1-Negative Patients

	H5N1-positive patients (n = 12)		H5N1-negative patients (n = 25)		
Feature	Mean value ± SD	Median value	Mean value ± SD	Median value	*P*
Age, year	6.7 ± 3.9	6	1.2 ± 2.9	0.3	<.001
pH	7.46 ± 0.07	7.49	7.29 ± 0.17	7.32	<.001
PaO_2, mmHg	61.4 ± 59.3	41.9	58.9 ± 23.8	58.6	.253
$PaCO_2$, mmHg	33.2 ± 12.4	32.5	47.5 ± 18.1	41.3	.009
FiO_2	0.81 ± 0.28	1	0.82 ± 0.27	1	.987
Body temperature at onset, °C	39.1 ± 0.4	39	37.7 ± 1	37.8	<.001
WBC count, cells/mm^3[a]	2863.6 ± 1545	2300	13,376 ± 9478	11,000	<.001
Platelet count × 10^3, cells/mm^3[a]	123.5 ± 52.3	125	366 ± 179.5	374	<.001
AST level, IU/L[a]	1723 ± 2784	724	259 ± 653	105	<.001
ALT level, IU/L[a]	628 ± 1042	248	221 ± 845	40	<.001

Variable	H5N1-positive patients, no. (%) of patients (n = 12)	H5N1-negative patients, no. (%) of patients (n = 25)	*P*
Prognosis			
Alive	2 (16.7)	13 (52.0)	.091
Dead	10 (83.3)	12 (48.0)	
Sex			
Male	8 (66.7)	8 (32.0)	.046
Female	4 (33.3)	17 (68.0)	
Multiple organ failure			
Yes	1 (8.3)	18 (72.0)	<.001
No	11 (91.7)	7 (28.0)	

NOTE. *P* values <.05 indicate statistically significant differences between H5N1-positive and H5N1-negative groups. ALT, alanine aminotransferase; AST, aspartate aminotransferase; SD, standard deviation; WBC, white blood cell.

[a] WBC and platelet counts were available for only 11 H5N1-positive patients, and AST and ALT levels were available for only 7 H5N1-positive patients and 23 H5N1-negative patients.

influenza virus infection and the identification of the key objective clinical data are crucial pieces of information that will help physicians provide timely and adequate treatment.

In the present study, we reviewed the clinical data from pediatric ARDS cases to identify distinctive findings of cases of H5N1 subtype influenza virus infection among Vietnamese children. This work was performed in close collaboration with the National Hospital of Pediatrics (NHP) in Hanoi and was supported by the Ministry of Health, Labour, and Welfare in Japan and the Ministry of Health in Vietnam.

MATERIALS AND METHODS

Data source. Clinical and laboratory data for pediatric patients (aged >1 month) with severe illness examined at the NHP from December 2003 through June 2008 were analyzed. Patients examined prior to 2007 were enrolled in the study retrospectively by hospital record review and were followed prospectively after hospital admission. The diagnosis of ARDS was made according to international standards [21], which involve acute onset; PaO_2/FiO_2 ratio (P/F ratio) <200, independent of controlled mechanical ventilation; and bilateral infiltration observed on chest radiography without left heart failure or with pulmonary artery wedge pressure <18 mmHg. We enrolled patients with severe ARDS whose P/F ratios were <100 during their clinical courses. H5N1 infection was confirmed with throat and/or nasal swabs tested by reverse-transcriptase polymerase chain reaction at the hospital laboratory or at the National Institute of Hygiene Epidemiology (Hanoi). The study was reviewed by the ethical committee of the International Medical Center Japan in 2007, and the design was approved on 28 September 2007.

Statistical methods. Fisher's exact test was employed for bivariate analysis of categorical data. The nonparametric Mann-Whitney test was used for 2-group comparisons of continuous data. Survival curves and rates were calculated by the Kaplan-Meier method. The log-rank (Mantel-Cox) test was used for the comparison of 2 survival curves. All statistical analyses were performed with SPSS, version 14.0 (SPSS).

RESULTS

Thirty-nine patients with ARDS who met the inclusion criteria visited the hospital during the study period, but 2 were excluded

Table 2. Summary of All Clinical Data

				Prognosis				Respiratory parameters				Liver function levels, IU/L		Blood counts[b]			
Patient	Sex	Age, years	Duration,[a] days	Death	MOF	BT at onset, °C	Blood pH	PaO_2, mmHg	$PaCO_2$, mmHg	P/F	Lowest P/F	AST	ALT	WBC	RBC	PLT	Cause of ARDS
H5N1 positive (n = 12)																	
1	F	12	7	Yes	Yes	39.5	7.48	29.1	30.7	29	29	NT	NT	2100	4510	45	Pneumonia (H5N1)
2	M	5	16	Yes	No	39.0	7.49	70.3	35.0	70	35	NT	NT	3400	4380	174	Pneumonia (H5N1)
3	M	10	11	Yes	No	39.5	7.51	33.5	28.1	84	24	NT	NT	2800	3750	143	Pneumonia (H5N1)
4	F	5	7	Yes	No	39.5	7.34	29.1	54.1	29	29	NT	NT	1100	3980	91	Pneumonia (H5N1)
5	M	4	15	Yes	No	39.5	7.5	40.9	26.1	41	36	NT	NT	2300	4190	150	Pneumonia (H5N1)
6	F	1	6	Yes	No	38.5	7.41	76.1	26.2	190	34	1121	484	NT	NT	NT	Pneumonia (H5N1)
7	M	1.3	9	Yes	No	39.0	7.52	243.0	9.3	607	65	8010	2972	1400	2190	31	Pneumonia (H5N1)
8	M	11	11	Yes	No	39.0	7.54	34.0	22.0	34	34	801	217	4800	4500	125	Pneumonia (H5N1)
9	F	9	26	No	No	39.5	7.47	42.8	34.2	43	43	312	248	6300	3970	122	Pneumonia (H5N1)
10	M	4	12	Yes	No	38.5	7.44	31.4	43.1	31	18	583	89	2300	4220	154	Pneumonia (H5N1)
11	M	7	18	No	No	38.5	7.49	58.0	41.0	116	97	511	105	3300	398	116	Pneumonia (H5N1)
12	M	11	10	No	No	39.5	7.55	52.0	29.6	52	49	724	282	1700	4530	207	Pneumonia (H5N1)
H5N1 negative (n = 25)																	
13	F	4	2	Yes	Yes	38.8	7.33	58.6	32.4	59	52	77	49.5	9300	2990	83	Pneumonia
14	M	0.2	23	No	Yes	34.3	7.35	34.5	41.3	35	31	142	67	3250	4030	57	Pneumonia
15	M	0.2	53	No	No	37.4	7.37	74.5	29.6	186	50	NT	NT	29200	4250	547	Unknown
16	F	3	29	No	Yes	38.5	7.19	25.0	50.0	25	25	55	36	4000	3030	37	Pneumonia
17	F	3	19	Yes	Yes	39.0	7.31	34.5	40.5	35	35	284	55	1600	3910	352	Pneumonia
18	M	0.4	2	Yes	Yes	38.5	7.32	108.0	28.2	108	47	120	31	4200	NT	332	Pneumonia
19	F	0.33	8	Yes	Yes	38.5	7.30	108.4	31.4	108	38	126	31	9500	3680	374	Pneumonia
20	F	0.18	10	Yes	Yes	37.2	7.29	67.0	43.0	67	31	105	37	19100	3690	448	Pneumonia (rhinovirus)
21	F	0.18	13	No	Yes	38.5	7.37	63.0	56.0	63	33	79	16	4200	3910	403	Pneumonia (rhinovirus)
22	F	0.18	6	Yes	Yes	37.5	7.31	24.0	56.5	24	24	77	44	23900	3640	534	Pneumonia (adenovirus)
23	F	0.25	8	Yes	Yes	38.0	7.23	42.7	75.7	43	26	287	51	16900	3450	176	Pneumonia (rhinovirus)
24	F	0.18	22	Yes	Yes	36.0	7.15	25.7	55.1	26	26	219	69	11000	3980	442	Pneumonia (rhinovirus)
25	F	0.25	5	Yes	Yes	37.8	6.61	75.6	113.6	76	76	196	40	14800	4010	590	Pneumonia
26	M	0.25	38	No	Yes	38.5	7.32	48.4	37.0	121	21	78	36	19000	4660	430	Pneumonia (bacterial)
27	F	14	12	No	Yes	37.5	7.45	95.1	37.0	95	95	3235	4097	28000	3510	278	Pneumonia
28	M	0.33	19	No	No	37.2	7.19	43.0	59.0	43	43	246	50	8600	3360	342	Pneumonia
29	M	0.25	36	No	Yes	36.9	7.32	39.0	40.0	98	49	83	25	1950	3200	497	Unknown
30	F	0.2	14	No	No	37.5	7.39	88.6	38.0	89	89	36	108	5300	5180	322	Pneumonia (bacterial)
31	F	0.12	35	No	Yes	37.2	7.41	61.0	41.0	153	55	118	52	13700	8480	592	Pneumonia
32	F	0.33	44	No	No	37.5	7.15	48.0	45.0	48	30	NT	NT	8800	4480	58	Pneumonia
33	F	0.2	35	No	Yes	38.5	7.30	70.0	31.0	175	49	99	38	28400	4290	532	Pneumonia
34	F	0.2	35	No	No	36.5	7.22	61.0	38.4	153	66	71	59	11400	3680	330	Pneumonia
35	M	0.25	22	Yes	Yes	38.0	7.47	58.0	63.0	73	22	46	14.6	10800	4140	508	Pneumonia (adenovirus)
36	F	0.2	23	Yes	No	38.6	7.36	65.0	50.0	65	27	111	27	12000	3440	233	Pneumonia
37	M	0.2	24	No	No	38.5	7.42	55.0	54.0	138	47	65	38	35500	3540	654	Unknown

NOTE. ALT, alanine aminotransferase; ARDS, acute respiratory distress syndrome; AST, aspartate aminotransferase; BT, body temperature; MOF, multiple organ failure; NT, not tested; P/F PaO_2/FiO_2 ratio; PLT, platelet; RBC, red blood cell; WBC, white blood cell.

[a] The duration represents the time from onset of illness to death or hospital discharge.

[b] WBC and RBC counts are cells/mm^3; PLT counts are 10^3 cells/mm^3.

because of insufficient medical data. Twelve of 37 patients were shown to be positive for H5N1 influenza by polymerase chain reaction performed in the laboratory of the NHP (table 1).

Most patients in the H5N1-positive group experienced rapid deterioration of ARDS and died of respiratory failure even with proper medical care. Although 3 patients had thrombocytopenia (patients 1, 4, and 7) and 7 (patients 6–12) had increased serum aminotransferase levels (table 2), multiple organ failure was not followed by pathological investigation for H5N1-positive patients (data not shown).

The P/F ratio of all patients enrolled in this study was <100 during their clinical courses. However, $PaCO_2$ level at hospital admission was lower among H5N1-positive than among the H5N1-negative patients, illustrating that ventilation capacity was higher in the H5N1-positive group, compared with the H5N1-negative group. These clinical features of ARDS in the H5N1-negative group made us wonder why ARDS in the H5N1-positive group was not more severe than that in the H5N1-negative group on hospital admission. The survival probability and days until final outcome (± standard deviation) among H5N1-positive ($n = 12$) and H5N1-negative ($n = 25$) patients were 17% and 52% and 12.3 ± 5.7 days (median, 11 days) and 21.5 ± 13.8 days (median, 22 days), respectively, demonstrating that the survival probability in the H5N1-positive group was significantly lower than that in the H5N1-negative group ($P = .022$, by log-rank test; $P = .038$, by Tarone-Ware test) (figure 1). These observations are the first step toward examining the clinical data for the H5N1-positive patients, which we designated as having fulminant ARDS.

As summarized in table 1, leukopenia and thrombocytopenia were observed in the H5N1-positive group, but leukophilia and normal-range thrombocyte levels were observed in the H5N1-negative group. Serum aspartate aminotransferase and alanine aminotransferase levels were also increased in the H5N1-positive but not the H5N1-negative group. Clinically, body temperature at illness onset in the H5N1-positive group was significantly higher than that in the negative group. We observed differences in clinical features between the H5N1-positive and H5N1-negative groups; we also analyzed differences with regard to sex, age distribution, and prognosis between these groups. The number of male patients in the H5N1-positive group was significantly higher than that in the H5N1-negative group, and H5N1-positive patients were significantly older than those in the H5N1-negative group. The mean time from illness onset until death (± standard deviation) was 10.4 ± 3.3 days (median, 10.5 days) in H5N1-positive group ($n = 10$) and 11.7 ± 3.3 days (median, 9 days) in H5N1-negative group ($n = 12$), and the mean time until hospital discharge (recovery) was 26 ± 18 days (median, 22 days) in the H5N1-positive group ($n = 2$) and 30.5 ± 11.8 days (median, 35 days) in the H5N1-negative group ($n = 13$). No significant differences were observed between groups with regard to time from illness onset to death.

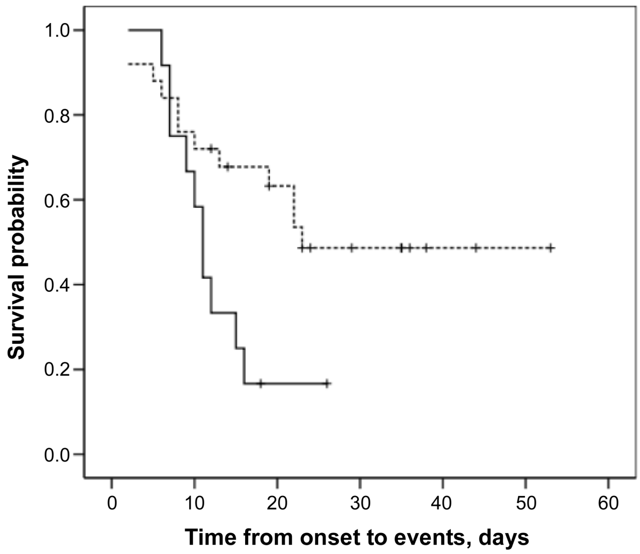

Figure 1. Survival probability for H5N1-positive (*solid line*) and H5N1-negative (*dashed line*) patients. For differences between 2 groups, $P = .022$ (by log-rank test) and $P = .038$ (Tarone-Ware test). The duration (± standard deviation) with disease until final outcome was 12.3 ± 5.7 days (median, 11 days) for the H5N1-positive group and 21.5 ± 13.8 days (median, 22 days) for the H5N1-negative group.

DISCUSSION

H5N1-infected patients were significantly older than patients in the comparator group. To our knowledge, the mortality rate by age has not been discussed precisely for pediatric patients with ARDS; Flori et al [22] collectively discussed age and mortality in their analysis of 328 patients with ARDS who were aged between 36 weeks (corrected gestational age) and 18 years, and the mortality rate among patients with a P/F ratio <100 was ~35%. The observed significant difference in mortality rate by age could prove that relatively older age (6.7 ± 3.9 vs 1.2 ± 2.9 years) is one of the risk factors for H5N1 infection.

ARDS frequently results in a lethal outcome attributable not only to respiratory failure but also to multiple organ failure [15, 23, 24]. Our study confirmed that survival of patients with ARDS aged <16 years is drastically improved by medical care, but that these patients die of respiratory failure and multiple organ failure [14]. On the other hand, most patients in the H5N1-positive group still showed rapid progress and deterioration of ARDS and died of respiratory failure, even with proper medical care followed by pathological investigation. Although aspartate aminotransferase and alanine aminotransferase levels were higher in the H5N1-positive but not the H5N1-negative group, the elevation of serum aminotransferase levels is a relatively common feature in H5N1 patients [6, 9, 25]. A review of 2 groups of patients in large case studies has shown that elevated aminotransferase levels are not thought to be specific in H5N1 patients [26, 27]. These observations strongly suggest that H5N1-infected patients die because of rapidly progressive respiratory failure before revealing typical multiple organ failure status accompanied by failure of multiple organs such as liver, heart, and kidney.

Physiologically, we further analyzed data regarding the P/F ratio, which is a good parameter of oxygenation capacity for respiratory function. P/F ratios were <100 in both H5N1-positive and -negative patients, which means that the oxygenation capacity in both groups was severely damaged. Surprisingly, $PaCO_2$ levels revealed that ventilation capacity was normal in H5N1-positive patients but was severely damaged in H5N1-negative patients with ARDS; that is, H5N1-negative patients experienced more severe respiratory failure than did H5N1-positive patients on hospital admission. The log-rank test and the more severely conditioned Tarone-Ware test also showed a significant difference in survival probability between the H5N1-positive and H5N1-negative groups. H5N1-positive patients started with normal ventilation capacity on hospital admission, then rapidly proceeded to severe respiratory failure and death. Patients in the H5N1-positive group demonstrated a shorter duration until final outcome than patients in the H5N1-negative group; therefore, we designated these as fulminant ARDS patients. The initial check of blood gas levels may be an early diagnostic indicator of H5N1 infection. There may also be some mechanisms that influence cell activity during H5N1 infection and accelerate alveolar damage, resulting in death [28, 29]. Pathology and immunomodulator activity in H5N1 infection have been discussed elsewhere [30], but precise mechanisms have not been clarified yet [31].

Body temperature was significantly higher in H5N1-positive patients at the onset of disease, compared with H5N1-negative patients. This seasonal influenza-like symptom appears early in the course of the disease, with a body temperature >38°C in almost all infected patients [26]. Significant leukopenia and thrombocytopenia were observed in the H5N1-positive group ($P < .001$). Leukopenia and thrombocytopenia are observed in the majority of patients with H5N1 [26, 27]. There has been some discussion of the possibility that lymphopenia and increased levels of lactate dehydrogenase at presentation are associated with a poor prognosis [27]. Further investigation into lymphopenia and liver function in H5N1 patients is necessary for clarification.

We have demonstrated here that H5N1 infection with ARDS starts with high fever but relatively mild respiratory symptoms, then proceeds to serious respiratory failure with lower survival probability and shorter periods of illness (fulminant ARDS), compared with ARDS without H5N1 infection. Leukopenia, thrombocytopenia, and liver function on hospital admission might be risk parameters and early indicators of patients with H5N1 influenza virus infection.

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