Respiratory syncytial virus (RSV) is the major cause of pediatric bronchiolitis and pneumonia. Most children experience an initial RSV infection within the first 2 years of life. In hospitalized infants <1 year of age, ~60% to 80% of lower respiratory infections are due to RSV. The mortality rate associated with RSV is twice that of the influenza virus during the first year of life, and its hospitalization rate is 3 times higher than that of the influenza virus within the first 5 years of life. Therefore, premature birth, bronchopulmonary dysplasia, congenital heart disease, and Down syndrome are risk factors for severe illness in RSV infection. For those high-risk infants, palivizumab prophylaxis has been strongly recommended, and this humanized monoclonal antibody has produced favorable results to date.

In children without underlying disease, the mortality rate is 3 times higher than that of the influenza virus within the first 5 years of life. Therefore, prematurity, bronchopulmonary dysplasia, congenital heart disease, and Down syndrome are risk factors for severe illness in RSV infection. For those high-risk infants, palivizumab prophylaxis has been strongly recommended, and this humanized monoclonal antibody has produced favorable results to date.

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usually develops within 1 to 2 days after the onset of clinical symptoms, such as high fever, cough, and fatigue. Although the mechanism of rapid progression of RSV-induced encephalopathy remains to be elucidated, cytokines and neurotrophins have been evaluated as prognostic markers.

In this report, we present the cases and autopsies of 2 children with sudden death related to RSV, both of whom were >1 year old, were not born prematurely, and were without underlying disease. Both died within half a day after symptom onset. Postmortem examinations revealed extensive bronchiolitis, severe brain edema, and extremely high serum levels of IL-6. These cases demonstrate that dramatic elevation of IL-6 may be correlated with sudden death in RSV infection.

**CASE 1**

The first case is a 19-month-old boy who was born at full-term and had developed normally. He had no medical history, such as asthma and pneumonia, and no familial history of immunodeficiency. He was the second child, and his 3-year brother had a 39°C fever one morning on October. Although the younger boy had a slight fever in the afternoon, he did not show prominent symptoms, such as a strong cough. However, the fever rose to >39°C at 7 PM, and he was given an acetaminophen suppository. He went to bed at 8 PM, and he was found in cardiopulmonary arrest in his bed at 10:30 PM. In an emergency hospital, he was intubated, and a total of 1.7 mg of epinephrine was administered, but resuscitation was unsuccessful. A chest radiograph on admission showed slight infiltrates in both hilar areas (Fig 1A). Subsequent computed tomography showed diffuse cerebral edema (Fig 1B).

A postmortem examination was performed to evaluate the cause of death. There was no sign of abuse. A rapid viral screen using nasopharyngeal secretions tested positive for RSV and negative for influenza. A blood neutralization test against RSV was weakly positive, and a complement fixation test was negative, indicating that the patient was clinically in the early phase of RSV infection (Table 1). Serum cytokine measurements demonstrated a markedly elevated IL-6 level, a moderate elevation of IL-8, and a normal C-reactive protein (CRP) level. Reverse transcription-polymerase chain reaction (RT-PCR) of lung tissue detected RSV-A. Quantitative Polymerase chain reaction (PCR) using the patient’s tissues showed remarkably high levels of IL-6 in the bronchi and brain (Fig 2). No major genetic mutation was detected in the Inherited Disease Panel (Thermo Fisher Scientific).

Histologic examination showed extensive bronchitis, with 56% of bronchioles obstructed with peeled epithelia and inflammatory cells (Fig 3A). Localized pneumonia was accompanied by neutrophils, lymphocytes, and macrophages (Fig 3B). The entire brain was severely edematous without inflammation (Fig 3C). Immunostaining showed that both main bronchi and 28% of the bronchioles were RSV positive (Fig 3D). In the lungs, some macrophages contained RSV, but only 6% of the alveolar epithelia were RSV positive (Fig 3E). In the whole brain, no RSV was observed, even...
in the areas of perivascular edema (Fig 3F). Degenerated astroglia, such as swollen bodies or cytoplasmic vacuoles, were not observed (Fig 3G).

**CASE 2**

The second case is a 19-month-old girl who was born at full-term and had developed normally. She had no medical history, such as asthma and pneumonia, and no familial history of immunodeficiency. She had no brothers or sisters. One day in December, she played in the preschool as always, where no epidemic was observed. She began to cough at 9 PM and was given tipepidine hibenzate. The cough continued and her temperature rose to 40°C at 11 PM. She was given acetaminophen, after which she fell asleep around midnight. After an additional hour and a half, she was found in cardiopulmonary arrest in her bed. Despite resuscitation using epinephrine, she did not recover at the emergency hospital. A chest radiograph on admission showed mildly increased opacity of both lung fields (Fig 1C). Subsequent computed tomography showed diffuse cerebral edema (Fig 1D).

To evaluate possible causes for the unknown and unexpected death, a postmortem examination was performed. There was no sign of abuse. A rapid viral screen using nasopharyngeal secretions was positive for RSV and negative for influenza. A blood neutralization test against RSV was weakly positive, and a complement fixation test was negative (Table 1). Serum cytokine measurements demonstrated a significantly elevated IL-6 level, a moderately elevated IL-8 level, and a slightly increased CRP level. IL-6 and IL-8 levels were also elevated in the cerebrospinal fluid. RT-PCR of lung tissue detected RSV-B. Quantitative PCR using the patient’s tissues showed remarkably high levels of IL-6 and IL-8 in the bronchi (Fig 2). No major genetic mutation was detected in the Inherited Disease Panel.

Histologic examinations showed extensive bronchitis (Fig 4A), with 38% of bronchioles obstructed with peeled epithelia and inflammatory cells. Mild pneumonia was mainly accompanied by lymphocytes (Fig 4B). The entire brain was so edematous that a slight subarachnoid hemorrhage was observed on the compressed cerebral sulcus (Fig 4C). Immunostaining showed that both main bronchi and 26% of the bronchioles were RSV positive (Fig 4D), but only 3% of alveoli were RSV positive (Fig 4E). In the brain, a few intravascular macrophages contained RSV (Fig 4F). Degradation of astroglia was not observed in the nearby white matter (Fig 4G).

**DISCUSSION**

Compared with the compromised infant with an underlying disease, the pathogenesis of RSV-induced sudden death in normally developed children remains to be elucidated. Metabolic disorders and cytokine storm have been assumed to trigger acute encephalopathy with multiple organ failure in some cases. Bronchial epithelial cells, alveolar epithelial cells, and peripheral blood mononuclear cells can produce IL-6 and IL-8 within 24 hours of RSV infection.
The fatal cases presented in this report demonstrated extensive bronchiolitis with airway obstruction and accompanying RSV infection, and their bronchial cytokine levels were extremely elevated. IL-6 and IL-8 can promote respiratory inflammation and induce acute respiratory distress syndrome.\textsuperscript{22,23} Although diffuse alveolar damage was not observed in our patients, it is possible that RSV-infected bronchial epithelia produced massive IL-6 and IL-8 in the incubation time, which aggravated the bronchiolitis and dyspnea in the early period of symptoms. Therefore, the high serum levels of IL-6 and IL-8 may reflect the severity of the acute respiratory failure.

Elevation of IL-6 and IL-8 in the cerebrospinal fluid has been reported in RSV-associated encephalopathy.\textsuperscript{13,24,25} The cerebral histopathology of the patients in this study showed severe edema damaging cortical neurons, but degradation of astroglia was not observed, which means that encephalopathy was just beginning. Because IL-6 and IL-8 increase vascular permeability,\textsuperscript{26,27} their rapid elevation may induce brain edema in the early phase of encephalopathy. We suspect that the airway obstruction of bronchiolitis and respiratory impairment with pneumonia could be the major cause of the collapse, and that severe brain edema may also contribute to cardiopulmonary arrest in the early phase of RSV infection.

A nationwide Japanese survey reported that 1.4% of hospitalized children <4 years of age die because of RSV infection, but most of them have underlying diseases.\textsuperscript{11} In this study, we first demonstrated the histopathological progression of RSV infection, resulting in sudden death in previously healthy children. These fulminant pathologic features developed within half a day of the onset of symptoms, after an incubation time of only a few days. The moderate neutrophil infiltration in Case 1 indicates that some bacterial superinfection worsened the pneumonia, but the viral infection was the main aggravating factor in both cases. The subclass of RSV was different in these cases, suggesting that prognostic determinants could be due to host factors rather than viral factors. However, the patients had no medical history of repeated infection, and major genetic mutations relating

\textbf{FIGURE 3}

to immunodeficiency were not detected. Therefore, the mechanism of susceptibility is unidentified. Some minor mutations or unknown immune response might be involved in these RSV-related sudden death cases.

Both cases presented in this report were given acetaminophen a few hours before cardiopulmonary arrest. Acetaminophen is the first-line choice for the treatment of fever and pain in children, but overdoses can cause acute liver failure. In our study, both cases were given the recommended dose. Furthermore, no pathologic changes were observed in their liver tissues, and their serum creatinine levels were in the normal range. Therefore, we consider the possibility of adverse events related to acetaminophen to be quite low.

In conclusion, extreme elevation of IL-6 and IL-8 developed early after symptom appearance. In particular, the IL-6 level was >200-fold higher than normal, suggesting that IL-6 elevation may predict the risk for RSV-related sudden death in previously healthy children.

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ABBREVIATIONS

CRP: C-reactive protein
PCR: polymerase chain reaction
RSV: respiratory syncytial virus
RT-PCR: reverse transcription-polymerase chain reaction

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