Impaired Pulmonary Function in Patients with Hemorrhagic Fever with Renal Syndrome

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Pulmonary and cardiac functions were investigated in 13 patients hospitalized with nephropathia epidemica, a European form of hemorrhagic fever with renal syndrome. As compared with reference values, the patients’ diffusion capacity for carbon monoxide was decreased (P = .002) and pulmonary clearance of inhaled technetium-99m-labeled diethylenetriamine pentaacetic acid was increased (P = .002). In four of 11 patients, arterial blood gas analysis disclosed a reduction in partial pressure of O₂ (<10 kPa) and oxygen saturation (<94%). In three of 13 patients, chest radiography revealed interstitial infiltrates or pleural effusions. Lung volumes and expiratory flow rates of the patients were not significantly changed. By electrocardiography and echocardiography, no significant cardiac dysfunction was demonstrable. The pulmonary dysfunction was best explained by an alveolocapillary lesion. The two hantavirus-caused clinical syndromes, hemorrhagic fever with renal syndrome and hantavirus pulmonary syndrome, may be pathophysiologically more similar than appears from the clinical presentations.

Hantaviruses are the causative agents of two clinical syndromes with different organ manifestations. Hemorrhagic fever with renal syndrome (HFRS) has been recognized for decades in Asia and Europe [1]. The renal affection is conspicuous with hematuria, proteinuria, oliguria followed by polyuria, and elevation of serum creatinine [2]. Several serotypes of hantaviruses are associated with HFRS, and the case fatality rate varies from <0.5% to 10%. Hantavirus pulmonary syndrome (HPS) was first observed in 1993 in the southwestern United States [3]. Although it is induced by a hantavirus closely related to the HFRS-associated agents, the clinical picture is strikingly different. Within the first week after onset, noncardiogenic pulmonary edema develops, and the fatality rate is ~50%.

This difference between HFRS and HPS in organ manifestations does not preclude an involvement of common pathophysiological mechanisms in the two illnesses. HFRS and HPS are both acquired by airway transmission and present with similar prodromes, including high fever, chills, headache, and abdominal and back pain [4]. Thrombocytopenia, coagulopathy, and plasma leakage occur in both conditions. In the European form of HFRS, nephropathia epidemica (NE), a pulmonary involvement, may occur more frequently than appears from the clinical presentation.

In the acute phase of the disease, pulmonary radiographic aberrations, including infiltrates and/or pleural effusions, have been demonstrated in 28%–53% of the patients [5, 6], and bronchoalveolar lavage analysis has disclosed an inflammatory response [7]. The possibility of shared pathophysiological denominators is also supported by occasional reports of noncardiogenic pulmonary edema in NE [8, 9] and the presence of some degree of renal dysfunction in cases of HPS [4, 10].

To further define the involvement of the lower respiratory tract in NE, we determined pulmonary and cardiac functions of patients in the acute phase of the disease. No such data seem to be available from previous studies of any form of HFRS. A question of fundamental interest was whether aberrations might be found that were suggestive of those of HPS.

Materials and Methods

Patients. The study included 13 patients (8 men and 5 women; median age, 44 years; range, 19–67 years) who were hospitalized 2–7 days (median, 5 days) following onset of fever. All patients had symptoms typical of NE. Each case was confirmed by indirect immunofluorescent assay findings of specific IgM and IgG antibodies against Puumala virus [11]. Three patients had been receiving long-term treatment with antihypertensive medication and one of them with antidepressive medication as well. The remaining patients were previously healthy. Four patients were smokers.

WBC and platelet counts, hematocrit, and hemoglobin concentration, as well as serum concentrations of creatinine, albumin, and C-reactive protein, were determined daily. Pulmonary and cardiac examinations were performed 0–4 days (median, 1 day) after admission to the hospital. Ten of 13 patients were examined at follow-up 2–24 months after discharge.

Electrocardiography. Standard 12-lead electrocardiography was performed with use of a Mingophone 7 (Siemens...
Elema, Stockholm). Patients with abnormal findings were reex-
amined before discharge and at follow-up.

_Echocardiography._ Standard transthoracic echocardiogra-
phy (sector, M-mode, and Doppler) was performed with use of
Vingmed CFM 750 or Vingmed CFM 800 (Vingmed, Horten,
Norway) by one of the authors (O.R.). Systolic and diastolic
left ventricular diameter, fraction shortening, and stroke vol-
ume were evaluated in relation to reference values [12]. When
technically possible, systolic right ventricular pressure was cal-
culated from the maximum pressure difference between the
ventricle and the atrium. The presence of pericardial fluid and
asnergy was investigated.

_Chest radiography._ Within 1–2 days after admission, con-
ventional chest radiography was performed with posteroanterior
and lateral projections, supplemented with horizontal pro-
jection, in order to detect pleural effusions.

_Arterial blood gas analysis._ Blood from the radial artery
of patients breathing room air was obtained on admission and
analyzed by routine hospital methods for partial pressure of O2
(PO2) (reference value, 10.0–13.0 kPa), PCO2 (4.6–6.0 kPa),
O2 saturation (94%–100%), pH (7.35–7.45), and base excess
(–3 to +3 mmol/L).

_Spirometry and diffusion capacity._ Lung volumes, expira-
tory flow rates, and diffusion capacity were determined by
computerized Jaeger equipment (Erich Jaeger GbMH, Würz-
burg, Germany). Forced expiratory volume in the first second
(FEV1) and maximum expiratory flow rate (MEF) at 75%, 50%,
and 25% of the forced vital capacity were determined from the
largest values of three flow-volume curves. Vital capacity (VC)
was measured as inspiratory VC. Total lung capacity (TLC)
was determined by a body-box method with use of a Jaeger
box. Alveolar volume (VA) was calculated as VA = TLC
– 0.15 L (anatomic dead space). Diffusion capacity for carbon
monoxide (DLCO) was measured as the transfer factor by a
single-breath method [13]. The transfer constant (KlCO) was
calculated as KlCO = DLCO/VA. Lung volumes, flow rates,
DLCO, and KlCO were expressed as percentage of predicted
values [13, 14].

_Pulmonary clearance of aerosolized technetium-99m-
labeled diethylenetriamine pentaacetic acid._ Technetium-
99m-labeled diethylenetriamine pentaacetic acid (99mTc-
DTPA) was prepared and administered as previously described
[15]. In brief, sodium pertechnetate (99mTcO4) was eluted from
a 99Mo/99mTc generator, DRN 4329 (Mal-
linckrodt Medical, Petten, The Netherlands). The 99mTc was
chelated to DTPA (Sorin SpA, Saluggia, Italy) by introduction
of 700 MBq of 99mTcO4 into a kit containing 10 mg of DTPA
in a maximum volume of 5 mL isotonic saline, yielding >97%
complete binding. The 99mTc-DTPA was prepared 1 hour before
use and stored in liquid nitrogen.

An aerosol of 700 MBq of 99mTc-DTPA was generated by
an ultrasound nebulizer (Variosonic, Rotkreuz, Switzerland).
Nebulizing rate was set at 2 mL/min. To optimize deposition
of aerosol in the periphery of the lungs, a settling bag of
10 L was interposed between the nebulizer and the mouthpiece.
Inhalation was performed in a sitting position for a period of
3 minutes. Immediately thereafter, the individuals were placed
in supine position under an Anger-type scintillation gamma
camera (General Electric, Horsholm, Denmark) with a large
field of view and a parallel-hole 140 keV collimator.

Dynamic acquisition of the detected lung field radioactivity
was recorded for 40 minutes. The acquisition was framed at
20-second intervals. Thirty minutes after the start of acquisi-
tion, a bolus of 30 MBq of 99mTc-DTPA was given intrave-
nously and flushed centrally with 10 mL of saline. Data were
processed by the drawing of separate regions of interest closely
around the lungs and the right ventricle of the heart.

The relative plasma volume of the lung to the heart (lung/
heart), as seen by the gamma camera, was calculated from
the immediate increase following the bolus injection of the
99mTc-DTPA. Time activity curves were generated for the lungs
[l(t)] and the heart [h(t)] for the period 0–30 minutes. Pulmo-
ary activity was corrected for plasma background by generat-
in the function L(t) = l(t) – [h(t) × lung/heart], where L(t)
is the time activity of the lungs, corrected for background. L(t)
was described by a monoeponential fit and characterized by
its half-life.

Pulmonary clearance of 99mTc-DTPA of the patients with
NE was compared with values of an internal reference group
at the Department of Clinical Physiology, consisting of 12
healthy, nonsmoking volunteers (median age, 24 years; range,
21–44 years).

_Statistical analyses._ Wilcoxon’s signed-rank test for paired
observations, the Mann-Whitney U test, and Spearman rank
correlation coefficient were used for statistical calculations.
All statistical tests were two-tailed.

_Results._

_Clinical data._ The median duration of hospitalization was
6 days (range, 4–9 days). The median blood hemoglobin con-
centration on admission was 143 g/L (range, 121–185). The
median maximum value of serum creatinine was 196 µmol/L
(range, 76–602); of C-reactive protein, 86 mg/L (range,
28–141); and of WBC counts, 8.2 × 10^9/L (range, 5.6–14.8).
The median minimum value of hematocrit was 38% (range,
31–47%); of platelet counts, 73\times 10^12/L (range, 5.6–14.8).

The median duration of hospitalization was 56 days (range,
21–81). The median blood hemoglobin concentration on
admission was 143 g/L (range, 121–185). The median max-
imum value of serum creatinine was 196 µmol/L (range,
76–602); and of platelet counts, 73\times 10^12/L (range, 5.6–14.8).

The median minimum value of hematocrit was 38% (range,
31–47%); of platelet counts, 73\times 10^12/L (range, 45–216); and
of serum albumin, 32.0 g/L (range, 26.2–36.9). Respiratory
symptoms (dry cough) and signs (diminished breath sounds)
were reported in three and two patients, respectively. On ad-
mission, two patients had systolic hypotension (80 and 90 mm Hg,
respectively). One of these patients was observed for a 12-hour
period in the intensive care unit. Parenteral fluid was given
to eight of 13 patients. Only two of them received >2 liters (total
volume, 3 and 4 L, respectively), indicating that overhydration
was not a major reason for our findings. No patient required
dialysis. All patients survived.
Electrocardiography. Six of 13 patients had transient, non-specific T-wave abnormalities, predominantly located in regions corresponding to the anterosetal and/or apical parts of the heart (leads V1–V4).

Echocardiography. In one of 13 patients, minimal pericardial effusion was noted (maximum, 1–2 mm). In two other patients a slightly increased diameter of the right ventricle was found, and a third patient had a slight elevation of the systolic right ventricular pressure (35 mm Hg).

Chest radiography. In three of 13 patients, chest radiography showed abnormalities. One patient had interstitial infiltrates in the lower part of the lung. Another patient had bilateral pleural effusions. In the third patient, a combination of these findings was demonstrated.

Arterial blood gas analysis. In 11 patients, arterial blood gas was analyzed. The median values of $P_\text{O}_2$ and $P_\text{CO}_2$ were 10.5 kPa (range, 8.0–13.6) and 5.0 kPa (range, 4.5–6.2), and the median value of $O_2$ saturation was 96.1% (range, 91.6%–98%). Four of 11 patients were hypoxemic ($P_\text{O}_2$: 8.0, 8.7, 8.8, and 9.5 kPa, respectively). In all 11 patients, pH and base excess values were within normal ranges.

Spirometry and diffusion capacity. VC, TLC, FEV$_1$, MEF$_{75\%}$, and MEF$_{50\%}$ values did not differ statistically from predicted values (figure 1). The mean MEF$_{25\%}$ value was 76.0% (range, 23.7%–182%) of the predicted value ($P = .023$). As compared with values recorded at follow-up, however, no significant difference was found ($P > .05$). In nine of 13 patients, DL$_{CO}$ and KL$_{CO}$ values were <80% of the predicted values. The mean of DL$_{CO}$ values as percentages of predicted values was 75.3% (range, 51.9%–100.7%), and that of KL$_{CO}$ was 69.2% (range, 40.9%–85.2%) ($P = .002$ and $P = .002$, respectively; figure 1).

Four patients were smokers. Even if these patients were omitted, the DL$_{CO}$ and KL$_{CO}$ values were significantly different from predicted values ($P = .011$ and $P = .008$, respectively). Those DL$_{CO}$ and KL$_{CO}$ values were also significantly lower than values obtained at follow-up ($P = .017$ and $P = .025$, respectively; figure 2). No statistically significant correlation was found between DL$_{CO}$ or KL$_{CO}$ values and maximum serum concentrations of creatinine or C-reactive protein, or minimum serum concentrations of albumin. Nor was there any correlation between DL$_{CO}$ or KL$_{CO}$ values and maximum hematocrit value, maximum WBC counts, or minimum platelet counts ($P > .05$).

Pulmonary clearance of aerosolized 99mTc-DTPA. Compared with the reference group, patients in the acute phase of NE had a significantly increased pulmonary clearance of 99mTc-DTPA ($P = .002$; figure 3). This was true even when the four smokers were excluded ($P = .018$). The pulmonary clearance of 99mTc-DTPA was significantly correlated to maximum WBC counts ($r = -.79$; $P = .009$) but not to maximum concentrations of serum creatinine or C-reactive protein or to minimum platelet counts ($P > .05$).

Discussion

In the acute phase of NE, pulmonary involvement was demonstrated in terms of reduced diffusion capacity and increased pulmonary clearance of 99mTc-DTPA. The findings were statistically significant even when smokers were excluded. Together with previous radiological [5, 6] and bronchoalveolar lavage [7] findings, the present data demonstrate convincingly the presence of pulmonary involvement in NE. It should be noted that the patients’ illness was relatively mild. Thus, pulmonary involvement seems to be a general trait of NE and not just an occasional finding in severe cases.

The present finding of pulmonary dysfunction in NE might seem to reconcile poorly with the general lack of overt respiratory symptoms in the disease. Impaired pulmonary function may, however, contribute to the state of general illness, including malaise, headache, and nausea. In fact, there are also reports of cases of NE with more obvious pulmonary symptoms, even noncardiogenic pulmonary edema [6, 9].

In the more severe form of HFRS caused by Hantaan virus, pulmonary changes have been reported in various phases of the disease. In the oliguric or diuretic phase [16], overt pulmonary edema was noted in about 2% of the patients. At autopsy, pulmonary edema was found in five of 17 patients dying in the preoliguric phase of the Hantaan virus–caused form of HFRS [8]. Altogether, pulmonary involvement may be quite
common in HFRS, although with a highly varying degree of clinical expression.

The mechanisms behind the pulmonary changes in NE are unknown. Basically, the lesions might be caused by viral infection per se and/or by the host response to the infection. From in vitro studies of cytopathogenicity, there is no conclusive evidence of a direct viral effect of hantavirus [17]. The information is not yet comprehensive, however, mostly because of the lack of suitable models of hantavirus infection. As opposed to direct viral cytotoxicity, the presence of host-derived events is lent support from reported data.

Most striking, bronchoalveolar lavage fluid of NE patients has shown inflammatory changes, including increased numbers of activated macrophages, natural killer cells, and CD8 T cells, as well as an increased concentration of fibronectin [7]. These findings may reflect both luminal and interstitial events and would be compatible with an antiviral host response resulting in epithelial cell injury and interstitial inflammation. The importance of host-derived events in the pathogenesis of HFRS is also supported by the coincidental appearance of clinical symptoms with humoral [11] and cell-mediated immune responses [18].

The vascular tissue may be involved in the genesis of the pulmonary changes of NE. Vascular dysfunction, expressed as impaired vascular tone and increased capillary permeability, is a characteristic feature of the more severe Hantaan virus–caused form of HFRS [19]. More specifically, the occurrence of vascular leakage is suggested by a combination of hemocoagulation and normal serum protein levels, as reported in the hypotensive phase of the disease [16].

In NE, the presence of radiologically demonstrable interstitial infiltrates and pleural effusions [5, 6] is compatible with increased permeability of pulmonary capillaries. In the present study, a decreased diffusion capacity was demonstrated, indicating a disturbed function of the alveolocapillary membrane. DL_{CO} and KL_{CO} values were equally decreased, suggesting that there was no overall reduction of the alveolar volume. The findings may reflect both luminal and interstitial events and would be compatible with an accumulation of fluid in the alveolocapillary region.

In this context, the previous demonstration of high, protracted plasma levels of TNF-α in the acute phase of NE [20] is of interest. TNF-α is one of several mediators known to induce increased capillary permeability [19, 21]. Another mediator molecule of interest is nitric oxide. Nitric oxide is an
Endothelial injury seems not to have been specifically studied with respect to the influence on pulmonary clearance of $^{99m}$Tc-DTPA. Hence, the present findings most probably reflected primary pulmonary events.

It is tempting to relate the present data not only to various forms of HFRS but also to HPS. The HPS-causing virus is closely related to Puumala virus [4]. HPS is clinically characterized by increased capillary permeability, mainly in the form of an excessive exudation of protein-rich fluid into the alveoli [3]. In HPS, respiratory distress and hypoxemia are usually prominent. In spite of the difference in severity, there are principal similarities in the pulmonary findings in HPS and HFRS.

In both conditions, early radiological findings include pleural effusions and interstitial infiltrates [5, 6, 29]. In similarity to HPS [30], and in contrast to the adult respiratory distress syndrome, only a modest accumulation of neutrophils occurs in the lower respiratory tract in the acute phase of NE [7]. Although in NE the pulmonary changes are usually too modest to lead to evident symptoms, they may obviously be similar by nature to those of HPS.

In conclusion, pulmonary function was substantially affected in patients with NE; these pulmonary changes could not be attributed to cardiac or renal dysfunction. Most probably, the pulmonary changes reflect alveolocapillary inflammation.

![Figure 3. Pulmonary clearance of technetium-99m-labelled diethylenetriamine pentaacetic acid ($^{99m}$Tc-DTPA) in 13 patients in the acute phase of nephropathia epidemica (NE) and in 12 healthy controls. Mean half-life ($T_{1/2}$) of radioactivity and 95% confidence intervals are indicated. The difference between the two groups was statistically significant ($P = .002$, Mann-Whitney $U$ test).](https://academic.oup.com/cid/article-abstract/25/5/1084/341284)

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