Clinical significance of interstitial lung disease and its acute exacerbation in microscopic polyangiitis

Word counts:

Abstract: 296

Text: 3660

Title: Clinical significance of interstitial lung disease and its acute exacerbation in microscopic polyangiitis

Running head: ILD and AE in MPA

Authors' full names:

Hironao Hozumi, M.D., Ph.D.[1]*, Masato Kono, M.D., Ph.D.[2], Hirotsugu Hasegawa, M.D., Ph.D.[3], Hideki Yasui, M.D., Ph.D.[1], Yuzo Suzuki, M.D., Ph.D.[1], Masato Karayama, M.D., Ph.D.[1], Kazuki Furuhashi, M.D., Ph.D.[1], Noriyuki Enomoto, M.D., Ph.D.[1]
Tomoyuki Fujisawa, M.D., Ph.D.[1], Naoki Inui, M.D., Ph.D.[1, 4], Yutaro Nakamura, M.D., Ph.D.[1], Koshi Yokomura, M.D., Ph.D.[2], Hidenori Nakamura, M.D., Ph.D.[3], Takafumi Suda, M.D., Ph.D.[1]

Authors' affiliation(s):

[1] Second Division, Department of Internal Medicine, Hamamatsu University School of Medicine, 1-20-1 Handayama Higashiku, Hamamatsu 431-3192, Japan

[2] Department of Respiratory Medicine, Seirei Hamamatsu General Hospital, 2-12-12 Sumiyoshi, Naka Ward, Hamamatsu, 430-8558, Japan

[3] Department of Respiratory Medicine, Seirei Mikatahara General Hospital, 3453 Mikatahara-cho, Kita-ku, Hamamatsu, 433-8558, Japan

[4] Department of Clinical Pharmacology and Therapeutics, Hamamatsu University School of

Medicine, 1-20-1 Handayama Higashiku, Hamamatsu 431-3192, Japan

*** Corresponding author:**

Hironao Hozumi

Second Division, Department of Internal Medicine, Hamamatsu University School of Medicine, 1-20-1 Handayama Higashiku, Hamamatsu 431-3192, Japan

E-mail address: hozumi@hama-med.ac.jp

ORCID ID: 0000-0001-5439-1543

Conflict of interest:

The authors declare no conflict of interests, including personal or financial support and no involvement with organization(s) that have financial interest in the subject matter.

Funding:

This work was supported by a grant from the Japan Society for the Promotion of Science (JP 18K15947 to H.H.).

Abbreviations list

AE, acute exacerbation; ANCA, antineutrophil cytoplasmic antibody; CS, corticosteroids; CTD, connective tissue disease; DAH, diffuse alveolar hemorrhage; $\%DL_{CO}$, percent predicted diffusing capacity of the lung carbon monoxide; EGPA, eosinophilic granulomatosis with polyangiitis; %FVC, percent predicted forced vital capacity; GPA, granulomatosis with polyangiitis; HRCT, high-resolution computed tomography; ILD, interstitial lung disease; IQR, interquartile range; IS, immunosuppressant; KL-6, Krebs von den Lungen-6; MPA, microscopic polyangiitis; MPO, myeloperoxidase; PaO_2, arterial

51 oxygen pressure; PR3, proteinase 3; RA, rheumatoid arthritis; SSc, systemic sclerosis; UIP,

52 usual interstitial pneumonia

Abstract

BACKGROUND: Presence of interstitial lung disease (ILD) is believed to be associated with mortality in microscopic polyangiitis (MPA); however, evidence on MPA-ILD remains lacking. Acute exacerbation (AE) refers to rapidly progressive, fatal respiratory deterioration that may develop in patients with various ILDs. No study has investigated the clinical significance of AE in MPA-ILD.

QUESTION: We aimed to determine the clinical picture and prognostic factors, the incidence of AE and its risk factors in patients with MPA-ILD.

STUDY DESIGN AND METHODS: Eighty-four consecutive patients with MPA-ILD and 95 patients with MPA-non-ILD were analyzed. We also compared 80 patients with MPA-ILD and 80 patients with idiopathic interstitial pneumonia without myeloperoxidase-anti-neutrophil cytoplasmic antibody-positivity (ILD-alone), who were matched for age, sex, and chest high-resolution computed tomography pattern.

RESULTS: The MPA-ILD group had a higher frequency of men and smokers and was associated with higher mortality than the MPA-non-ILD group. The matched MPA-ILD group had a higher mortality rate than the matched ILD-alone group. There was no significant difference in AE incidence between the matched MPA-ILD and ILD-alone groups (1-year AE cumulative incidence rate: 7.5% and 5.2%, respectively, $P = 0.75$). In the MPA-ILD group, a lower percent-predicted forced vital capacity (%FVC) was independently associated with a higher mortality rate (hazard ratio [HR]: 0.96 per 1% increase, $P < 0.01$) and a higher AE incidence rate (HR: 0.96 per 1% increase, $P = 0.01$). On multivariable Cox regression analysis with time-dependent covariates, developing AE during their clinical course was strongly associated with shorter survival (HR: 17.1, $P < 0.001$).

INTERPRETATION: MPA-ILD represented a distinct phenotype with poor prognosis. Lower %FVC was an independent prognostic factor. Patients with lower %FVC had a risk of

developing AE, which was a strong prognostic determinant. The specific management for MPA-ILD and AE should be established.

Microscopic polyangiitis (MPA) is a systemic, pauci-immune, necrotizing vasculitis that primarily affects small vessels, which is associated with antineutrophil cytoplasmic antibodies (ANCAs) [1,2]. MPA belongs to rare disease entities of ANCA-associated vasculitides (AAVs), including granulomatosis with polyangiitis (GPA) and eosinophilic GPA (EGPA) [3-5]. Most of the patients with MPA are seropositive for ANCA reacting with myeloperoxidase (MPO-ANCA), and only a few of the patients react with proteinase 3 (PR3-ANCA). Reportedly, MPO-ANCA-positive AAV and PR3-ANCA-positive AAV are distinct autoimmune syndromes with different genetic backgrounds [6,7], and MPO-ANCA, rather than PR3-ANCA, plays a role in the pathogenesis of MPA [8]. Patients with MPA can present various organ involvements, including the kidneys, lungs, skin, and nerves [9]. Interstitial lung disease (ILD) is detected in 10%–60% of patients with MPA, which is associated with mortality when present [10-14]. However, current evidence on MPA-ILD was largely based on small cohort studies/case series and studies analyzing MPA, GPA, and EGPA together as a group of AAV. Therefore, the clinical picture of the phenotype of "MPA-ILD" is yet to be fully elucidated.

Acute exacerbation (AE) is a rapidly progressive, fatal respiratory deterioration that develops unpredictably during the clinical course of IPF [15,16]. AE also occurs in patients with various ILDs other than IPF, including connective tissue disease-associated ILDs (CTD-ILDs) [17-19]. However, the evidence on AE in ILDs other than IPF is still lacking. The precise AE incidence rate, its predictive factors, and its impact on prognosis in patients with MPA-ILD remain unknown. Here, this multicenter, retrospective cohort study was conducted to determine the clinical significance of ILD and its AE in patients with MPO-ANCA-positive MPA.

Methods

Patients and diagnostic criteria:

We retrospectively screened 513 consecutive patients who had had MPO-ANCA seropositivity between 2007 and 2019 at the Hamamatsu University, Seirei Mikatahara General, or Seirei Hamamatsu General Hospitals (Figure 1A). Of them, 218 patients were classified as MPA based on the European Medicines Agency algorithm and the Chapel Hill Consensus Conference criteria, with a consensus among rheumatologists, pathologists, and attending physicians [1,2,20]. Therefore, patients with isolated MPO-ANCA positivity and ILD were not included in this study [21,22]. Patients with no available high-resolution computed tomography (HRCT) data at the time of MPA diagnosis; initial presentation of AE or diffuse alveolar hemorrhage (DAH); the presence of advanced malignancy at baseline; a CTD at baseline; insufficient baseline data (e.g., percent-predicted forced vital capacity [%FVC]); and MPO-ANCA and PR3-ANCA double-positive MPA were excluded. Subsequently, we classified patients into those with ILD and those without, based on the assessment of chest HRCT at the time of MPA diagnosis. Consequently, 84 patients with ILD (MPA-ILD group) and 95 without ILD (MPA-non-ILD group) were enrolled and compared. In addition, we reviewed 253 patients with idiopathic interstitial pneumonia diagnosed between 2007 and 2019 at the Hamamatsu University hospital, without the development of MPA or connective tissue disease, and who were negative for MPO-ANCA (ILD-alone) (Figure 1B). We then selected and compared 80 patients with MPA-ILD and 80 patients with ILD-alone who were propensity score-matched for age, sex, and high-resolution computed tomography pattern [usual interstitial pneumonia (UIP) or other than UIP (non-UIP)] (e-appendix 1).

DAH was diagnosed based on the following criteria: 1) diffuse ground-glass opacity and/or consolidation on HRCT without alternative explanation and 2) hemoptysis, bronchoscopic evidence of overt hemorrhage, or bloody returns from the bronchoalveolar lavage (BAL) fluid [14,23,24]. AE of MPA-ILD was defined based on the 2016 International

Working Group report for AE-IPF, with slight modifications [15], as events meeting all the following criteria: 1) previous or concurrent diagnosis of MPA-ILD; 2) acute worsening or development of dyspnea typically within a 1-month duration; 3) HRCT with new bilateral ground-glass opacity and/or consolidation superimposed on a background reticular opacities/honeycombing; 4) deterioration not fully explained by cardiac failure or fluid overload; and 5) ruling out DAH by BAL.

Serum MPO-ANCA levels at the time of MPA diagnosis were measured using enzyme-linked immunosorbent assay kits or chemiluminescent enzyme immunoassay kits. In this study, MPO-ANCA levels were expressed as the ratio of MPO-ANCA titer to cut-off level because the cut-off levels differ according to the kits used in each hospital.

Baseline data pertaining to the following variables were collected from the medical records: clinical data; pathological findings; laboratory data, including serum MPO-ANCA titer, C-reactive protein (CRP), Krebs von den Lungen-6 (KL-6), and arterial oxygen pressure (PaO_2); results of pulmonary function tests, including %FVC and predicted diffusing capacity of the lung carbon monoxide ($\%DL_{CO}$); HRCT; MPA-related manifestation/involvement, including general symptoms (e.g., fever and arthralgia); the 1996 five-factor score [25]; treatment; and outcomes.

This multicenter study was conducted in accordance with the Declaration of Helsinki and was approved by the institutional review board of each participating institution (Hamamatsu University School of Medicine [approval number: 19-206], Seirei Mikatahara General Hospital [approval number: 19-42], and Seirei Hamamatsu General Hospital [approval number: 3211]). Written informed consent was not required because of the retrospective nature of the study.

HRCT assessment:

Chest HRCT images were reviewed by radiologists. ILD was defined as the evidence of bilateral reticular opacities with/without traction bronchiectasis on HRCT. In patients with ILD, HRCT patterns were further classified based on the 2018 IPF guidelines as UIP, probable UIP, indeterminate for UIP, and alternative diagnosis patterns[16].

Statistical methods:

Data were expressed as means (± standard deviation), median [interquartile range (IQR)], or frequency (%). Fisher's exact test or chi-square test was used for comparing proportions among groups. Between-group differences were assessed using Welch's unequal variances t-test. Correlation between different variables was evaluated using Spearman's correlation test. The observation period lasted from the date of MPA diagnosis until the last visit (the date of censoring or the date of death). AE-free survival was calculated from the date of MPA diagnosis until the date of AE onset in patients who developed AE or until the last visit in those without AE. The cumulative survival and AE incidence rates were calculated using the Kaplan–Meier test; the log-rank test and Gray's test (treating death as a competing event) were used to assess between-group differences. Cox proportional hazards regression analysis (with/without time-dependent covariates) was used to identify the prognostic factors and subdistribution hazard analyses were performed, according to the method of Fine and Gray, to identify the predictive factors of AE development (treating death as a competing event) and ILD-related death (treating other causes of death aside from those related to ILD as a competing event); age, sex, treatment, and all variables that showed a significant association in the univariate analysis were included in the multivariable analysis. We considered all *P*-values < 0.05 as indicating statistical significance. We analyzed all data using EZR (Jichi Medical University, Saitama, Japan), which is a graphical user interface for R (The R Foundation for Statistical Computing, Vienna, Austria).

Results

MPA-ILD vs. MPA-non-ILD:

The patient characteristics are summarized in Table 1. The MPA-ILD group showed significantly higher proportions of men and former/current smokers than the MPA-non-ILD group. Regarding the manifestations/involvements other than pulmonary involvement, the MPA-ILD group had a significantly lower prevalence of general symptoms and renal involvement than the MPA-non-ILD group.

Out of 84 patients in the MPA-ILD group, ILD diagnosis preceded the clinical onset of MPA in 42 (50%) patients (ILD-preceding group), and the median time from ILD diagnosis until MPA diagnosis was 39.1 (interquartile range: 18.3–83.5) months. Of the 42 patients in the ILD-preceding group, 15 (35.7%) were MPO-ANCA-positive at the time of ILD diagnosis, 12 (28.6%) were MPO-ANCA-negative at the time of ILD diagnosis but later had positive conversion, and 15 (35.7%) had MPO-ANCA results unknown at the time of ILD diagnosis but were MPO-ANCA-positive at the time of MPA diagnosis. Of the ILD-preceding group, 8 patients underwent surgical lung biopsy before the clinical onset of MPA (e-Table 1). In the remaining 42 (50%) patients other than the ILD-preceding group, the presence of ILD was found at the time of MPA diagnosis. The proportion of patients who received corticosteroids (CS) plus an immunosuppressant (IS) tended to be relatively higher in the MPA-ILD group than in the MPA-non-ILD group. ILD-related events, such as AE and chronic respiratory failure, were the most common causes of death in the MPA-ILD group, while infection was the major cause of death in the MPA-non-ILD group. The cumulative survival rate was significantly lower in the MPA-ILD group than in the MPA-non-ILD group (Figure 2A).

MPA-ILD vs. ILD-alone:

The patient characteristics are summarized in Table 2. At baseline, the matched MPA-ILD group had higher %FVC than the matched ILD-alone group. The matched MPA-ILD group had a greater proportion of patients receiving immunosuppressive therapy than the matched ILD-alone group. The matched MPA-ILD group had a significantly lower cumulative survival rate than the matched ILD-alone group (Figure 2B); however, there was no significant between-group difference with respect to the AE incidence rate (Figure 2C). The incidence of ILD-related or other death is shown in e-Figure 1. There was no significant difference in the incidence of ILD-related death between the matched MPA-ILD and the matched ILD-alone groups (e-Figure 1A). However, the incidence of death from causes other than ILD-related events was higher in the matched MPA-ILD group than in the matched ILD-alone group (5-year cumulative incidence rate: 34.0% vs. 14.2%, respectively, $P < 0.001$) (e-Figure 1B). ILD-related events were the most common causes of death in both groups. However, more patients in the matched MPA-ILD group died due to infection, bleeding-related events, or uremia.

Prognostic factors of MPA-ILD:

The results of Cox proportional hazards analysis of all-cause mortality are presented in Table 3. In the multivariable analysis, lower %FVC was independently associated with increased all-cause mortality (hazard ratio [HR]: 0.96 per 1% increase, $P < 0.01$).

The results of the subdistribution hazards analysis of ILD-related death are presented in e-Table 2. In the multivariable analysis, higher KL-6 levels were independently associated with ILD-related death (HR: 1.07 per 100 U/mL increase, $P < 0.01$) (model 1). KL-6 levels showed a significant negative correlation with %FVC (correlation coefficient: −0.33, $P < 0.01$). Therefore, to avoid multicollinearity, we performed two separate models of

multivariable analyses that incorporated "age, sex, induction treatment regimen, and KL-6 levels" (model 2), or "age, sex, induction treatment regimen, and %FVC" (model 3). In models 2 and 3, higher KL-6 levels and lower %FVC were independently associated with ILD-related death, respectively.

Analysis on AE in MPA-ILD:

In the entire MPA-ILD group, the 1-year AE cumulative incidence rate was 7.2%. BAL fluid findings at AE onset are presented in e-Table 3. The results of the subdistribution hazards analysis of AE development are presented in e-Table 4. In the multivariable analysis, lower %FVC was independently associated with a higher AE incidence rate (HR: 0.96 per 1% increase, $P = 0.01$).

Cox proportional hazards analysis with time-dependent covariates that were adjusted for age, sex, induction treatment regimen, and %FVC were performed to clarify the association between AE development, a time-dependent covariate, and mortality. In this model, the development of AE during the clinical course of MPA-ILD was significantly associated with increased mortality (HR: 17.1, 95% confidence interval: 6.04–48.4, $P < 0.01$). All patients who developed AE were treated with high-dose CS. The median survival time from AE onset was 0.76 months, and the post-AE 3-month survival rate was 25% (Figure 3).

Sub-Analysis:

The survival and AE incidence rates of patients with MPA-ILD by HRCT patterns are shown in e-Figure 2. The HRCT-UIP pattern group tended to have a lower survival rate than the HRCT-non-UIP pattern group (5-year cumulative survival rate: 35.8% vs. 54.2%, respectively, P = 0.18) (e-Figure 2A). The HRCT-UIP pattern group tended to have a higher AE incidence rate than the HRCT-non-UIP pattern group (3-year cumulative AE incidence rate; 20.2% vs.

13.1%, respectively, P = 0.17) (e-Figure 2B).

The frequency of MPA relapse with DAH is shown in e-Figure 3, which was higher in the MPA-ILD group than in the MPA-non-ILD group (1-year cumulative incidence rate: 8.4% vs. 1.1%, respectively; $P < 0.001$). The results of the subdistribution hazards analysis of MPA relapse with DAH are presented in e-Table 5; no predictive factors were identified. Cox proportional hazards analyses with time-dependent covariates that were adjusted for age, sex, induction treatment regimen, and %FVC were performed to clarify the association between MPA relapse with DAH and mortality. MPA relapse with DAH was significantly associated with increased mortality (HR: 3.02, 95% confidence interval: 1.03–8.83, $P = 0.04$).

Discussion

To our knowledge, this is the first and largest study that clarified the clinical significance of ILD and its AE in patients with MPA, based on the clinical, radiological, and physiological data. In the present study, the MPA-ILD group showed higher proportions of men and smokers than the MPA-non-ILD group. The MPA-ILD group had a significantly poorer prognosis with significantly different causes of death than the MPA-non-ILD group or the age-, sex, and HRCT pattern-matched ILD-alone group. In patients with MPA-ILD, lower %FVC was independently associated with increased mortality. Interestingly, AE did occur in patients with MPA-ILD, and the 1-year AE cumulative incidence rate was 7.2%. Lower %FVC was an independent predictor of AE. Of note, multivariable analysis with time-dependent covariates demonstrated that developing AE during the clinical course of MPA-ILD was strongly associated with a poor prognosis.

Among patients with MPA, the prevalence of ILD was not fully established; however, it seems to be higher in Asia (37%–60%) than in Europe (10%–39%) [10,12,26-28]. Consistent with these reports, the present study indicated the relatively high coexistence of

ILD (46.9%) in our series of MPO-ANCA-positive MPA patients. Interestingly, half of the patients with MPA-ILD in this cohort were diagnosed with ILD before the clinical onset of MPA, and the other half had ILD at the time of MPA diagnosis, suggesting that ILD may precede the clinical onset of MPA in most, but not all, patients with MPA-ILD.

The cumulative survival rate in patients with MPA is variable across study cohorts. Reportedly, the 5-year survival rate in those patients was 46%–80% [29], especially in those with ILD, 29%–60% [30]. However, recent studies have shown a gradual improvement in the survival rate of AAV including MPA over the last few decades [12,31]. In the present study, the cumulative survival rate in patients with MPA was relatively low, which may be due to the fact that approximately 30% of the patients received a combination of CS and IS therapy and the relatively high mean age of patients in this cohort.

To date, a few studies have compared the clinical characteristics between patients with ILD and those without ILD in MPA. The present study found that the proportions of men and smokers and the prevalence of general symptoms and renal involvement were significantly different between the MPA-ILD group and the MPA-non-ILD group. In accordance with our observations, several studies have shown the predominance of men and smokers in the MPA-ILD group [10,11,13,27]. The present study clearly indicated a large prognostic difference between the MPA-ILD group and MPA-non-ILD group, which is consistent with the results of previous studies [10,12,28]. We also found a significant difference in the causes of death between the two groups. ILD-related conditions were the most common causes of death in the MPA-ILD group, whereas infection was the major cause in the MPA-non-ILD group. These observations suggest that, among MPA, MPA-ILD is a distinct phenotype with poor prognosis, and specific management may be required for MPA-ILD. Anti-fibrotic agents, pirfenidone and nintedanib, has recently been approved for patients with IPF [32,33]. More recently, two randomized studies demonstrated the effectiveness of nintedanib

in progressive fibrosing ILDs, such as systemic sclerosis-associated ILD (SSc-ILD) and rheumatoid arthritis-associated ILD (RA-ILD), although these studies did not include MPA-ILD [34,35]. Thus, anti-fibrotic drugs may be a promising therapeutic agent for MPA-ILD [34-36].

To the best of our knowledge, this is the first study that compared a relatively large cohort of age-, sex-, and HRCT pattern-matched pairs of patients with MPA-ILD and those with ILD-alone. Notably, the matched patients with MPA-ILD showed a significantly poorer prognosis, despite better respiratory function than those with ILD-alone. While the number and incidence of ILD-related death were similar in both groups, the MPA-ILD group had a greater number of bleeding-related deaths, renal-related deaths, and infection-related deaths; moreover, the incidence of death from causes other than ILD-related was also significantly higher in the MPA-ILD group. These results suggest that matched patients with MPA-ILD and those with ILD-alone have a similar risk of ILD-related mortality; however, those with MPA-ILD have a poorer prognosis owing to the additional risk of death from non-ILD complications as compared to patients with ILD-alone.

In patients with MPA, older age and having ILD were associated with mortality [12,28]; however, no study has identified the prognostic factors in patients with MPA-ILD. Using a multivariable analysis, the present study, for the first time, demonstrated that lower %FVC was an independent prognostic factor in patients with MPA-ILD. Among other ILDs, such as IPF, SSc-ILD, RA-ILD, polymyositis/dermatomyositis-associated ILD, and Sjögren's syndrome-associated ILD, lower baseline %FVC was a major prognostic factor [37-41]. Thus, impaired lung function is likely to be a universal prognostic factor in various types of ILDs, including MPA-ILD.

AE is a life-threatening event in patients with IPF. However, growing evidence, including ours, indicated that patients with ILD other than IPF also develop AE during their

clinical course [17-19]. Regardless of the types and/or causes of ILD, patients developing AE exhibit an extremely poor prognosis with high mortality [42]. Interestingly, the present study showed that patients with MPA-ILD also developed AE, and the 1-year AE cumulative incidence rate was 7%. The reported annual AE incidence rates were 5%–15%, 2%–4%, and 2%–3% in IPF, idiopathic nonspecific pneumonia (NSIP), and RA-ILD, respectively [15,17-19]. Thus, the incidence of AE in MPA-ILD was similar to that reported in IPF and higher than that reported in idiopathic NSIP, and that reported in RA-ILD. In IPF, AE is more common in patients with physiologically and functionally advanced disease; lower baseline %FVC is the most consistent risk factor for AE-IPF [15]. Similar to IPF, lower %FVC was an independent predictor of AE in patients with MPA-ILD. The outcome of AE in MPA-ILD was extremely poor. Moreover, Cox proportional hazards analyses with time-dependent covariates showed that AE development was significantly associated with increased mortality. Taken together, more attention should be paid to the developing AE during the clinical course in patients with MPA-ILD, especially if the patients have a lower lung function.

Although UIP pattern on HRCT is the hallmark radiologic pattern for IPF [16], it is often seen also in patients with ILDs other than IPF, including MPA-ILD [10,27]. We previously reported that UIP pattern on HRCT was associated with AE development and poor prognosis in patients with RA-ILD [19]. It was also suggested that UIP pattern on HRCT may be associated with an increased risk of developing AE in various ILDs [42,43]. A recent study demonstrated that UIP pattern on HRCT was a prognostic factor in patients with AAV-ILD, including MPA and GPA [13]. In this study, although UIP pattern on HRCT tended to be associated with higher AE incidence and poorer prognosis, the differences did not reach statistical significance. It is possible that the sample size and number of events, including AE and death, may not have been sufficient to support our hypothesis. However, the impact of the UIP pattern on the incidence of AE and mortality may vary depending on the underlying

disease. Further studies are needed to clarify the clinical implication of UIP pattern on HRCT in MPA-ILD.

DAH is a serious complication in patients with AAV, which may occur as an initial presentation or as disease relapse [24]. In the present study, we focused on DAH as disease relapse, which occurred after successful induction treatment. Remarkably, the MPA-ILD group had a relapse with DAH more frequently than the MPA-non-ILD group. The reason for this remains unclear. It is hypothesized that a chronic subclinical alveolar hemorrhage is associated with lung fibrosis [44]. Thus, the preexisting subclinical hemorrhage in MPA-ILD may be partially attributable to this. Collectively, these observations emphasize the need for caution in the higher incidence of disease relapse with DAH in patients with MPA-ILD.

The present study had several limitations. Firstly, the retrospective design of the study renders it vulnerable to several biases. Our institutions are regional ILD referral centers, which may have introduced an element of selection bias. A prospective study is required to validate our results. Secondly, this study only included patients with MPO-ANCA-positive MPA. However, studies suggested that PR3-ANCA-positive AAV and MPO-ANCA-positive AAV are distinct syndromes [6,7]. Therefore, a different study design is needed to analyze PR3-ANCA-positive patients. Thirdly, in our study, it was not mandatory to perform a BAL for the diagnosis of DAH. Therefore, the incidence of DAH may have been underestimated. Fourthly, there were several patients who could not be diagnosed with either AE or DAH because of severe acute respiratory failure that prevented us from performing a BAL. The presence of such patients may have affected the incidence of AE and DAH and the results of risk factor analyses. Fifthly, in the MPA-non-ILD group, chest HRCT was performed at the time of MPA diagnosis; however, chest HRCT was not routinely performed after MPA diagnosis. Therefore, we could not fully ascertain whether patients in the MPA-non-ILD group developed ILD after the diagnosis of MPA. Finally, the different treatment regimens

may have affected the outcomes in our study population.

Interpretation

The present study demonstrated that patients with MPA-ILD had different characteristics and outcomes from those with MPA-non-ILD. Patients with MPA-ILD showed worse prognosis than those with MPA-non-ILD or those with ILD-alone, and lower %FVC was a poor prognostic factor. During the clinical course of MPA-ILD, AE did occur with high mortality, and lower %FVC was an independent predictor of this devastating condition. In the treatment of MPA, rheumatologists and pulmonologists should be aware of the poor prognostic significance of concurrent ILD and its AE, especially if the patients had lower %FVC. Establishment of specific management for MPA-ILD and AE is urgently needed.

Acknowledgments

Guarantor statement:

H.H. had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Author contributions:

N.A., H.H., N.E., T.F., N.I., Y.N., and T.S. designed the research; N.A., H.H., T.I., J.O., K.S., H.Y., Y.S., M.K., M.K., K.F., N.E., T.F., N.I., Y.N., and T.S. contributed to the acquisition or analysis of the data; N, A. and H.H. wrote the initial and final drafts of the manuscript; N.A., H.H., T.I., J.O., K.S., H.Y., Y.S., M.K., M.K., K.F., N.E., T.F., N.I., Y.N., and T.S. revised the drafts of the manuscript; and all authors approved the final version of the manuscript.

Funding:

405 This work was supported by a grant from the Japan Society for the Promotion of Science (JP
406 18K15947 to H.H.). The funding source did not provide any input or contributions in the
407 development of the research or manuscript.

408

References

1. Jennette JC, Falk RJ, Bacon PA, et al. 2012 revised International Chapel Hill Consensus Conference Nomenclature of Vasculitides. *Arthritis and rheumatism.* 2013;65(1):1-11.
2. Watts R, Lane S, Hanslik T, et al. Development and validation of a consensus methodology for the classification of the ANCA-associated vasculitides and polyarteritis nodosa for epidemiological studies. *Annals of the rheumatic diseases.* 2007;66(2):222-227.
3. Lane SE, Watts RA, Shepstone L, Scott DG. Primary systemic vasculitis: clinical features and mortality. *QJM : monthly journal of the Association of Physicians.* 2005;98(2):97-111.
4. Mukhtyar C, Flossmann O, Hellmich B, et al. Outcomes from studies of antineutrophil cytoplasm antibody associated vasculitis: a systematic review by the European League Against Rheumatism systemic vasculitis task force. *Annals of the rheumatic diseases.* 2008;67(7):1004-1010.
5. Jardel S, Puechal X, Le Quellec A, et al. Mortality in systemic necrotizing vasculitides: A retrospective analysis of the French Vasculitis Study Group registry. *Autoimmunity reviews.* 2018;17(7):653-659.
6. Lyons PA, Rayner TF, Trivedi S, et al. Genetically distinct subsets within ANCA-associated vasculitis. *The New England journal of medicine.* 2012;367(3):214-223.
7. Cornec D, Cornec-Le Gall E, Fervenza FC, Specks U. ANCA-associated vasculitis - clinical utility of using ANCA specificity to classify patients. *Nature reviews. Rheumatology.* 2016;12(10):570-579.
8. Chen M, Kallenberg CG. ANCA-associated vasculitides--advances in pathogenesis

and treatment. *Nature reviews. Rheumatology.* 2010;6(11):653-664.

9. Greco A, De Virgilio A, Rizzo MI, et al. Microscopic polyangiitis: Advances in diagnostic and therapeutic approaches. *Autoimmunity reviews.* 2015;14(9):837-844.

10. Tzelepis GE, Kokosi M, Tzioufas A, et al. Prevalence and outcome of pulmonary fibrosis in microscopic polyangiitis. *The European respiratory journal.* 2010;36(1):116-121.

11. Arulkumaran N, Periselneris N, Gaskin G, et al. Interstitial lung disease and ANCA-associated vasculitis: a retrospective observational cohort study. *Rheumatology (Oxford, England).* 2011;50(11):2035-2043.

12. Schirmer JH, Wright MN, Vonthein R, et al. Clinical presentation and long-term outcome of 144 patients with microscopic polyangiitis in a monocentric German cohort. *Rheumatology (Oxford, England).* 2016;55(1):71-79.

13. Maillet T, Goletto T, Beltramo G, et al. Usual interstitial pneumonia in ANCA-associated vasculitis: A poor prognostic factor. *Journal of autoimmunity.* 2020;106:102338.

14. Yamagata M, Ikeda K, Tsushima K, et al. Prevalence and Responsiveness to Treatment of Lung Abnormalities on Chest Computed Tomography in Patients With Microscopic Polyangiitis: A Multicenter, Longitudinal, Retrospective Study of One Hundred Fifty Consecutive Hospital-Based Japanese Patients. *Arthritis & rheumatology (Hoboken, N.J.).* 2016;68(3):713-723.

15. Collard HR, Ryerson CJ, Corte TJ, et al. Acute Exacerbation of Idiopathic Pulmonary Fibrosis. An International Working Group Report. *American journal of respiratory and critical care medicine.* 2016;194(3):265-275.

16. Raghu G, Remy-Jardin M, Myers JL, et al. Diagnosis of Idiopathic Pulmonary Fibrosis. An Official ATS/ERS/JRS/ALAT Clinical Practice Guideline. *American*

journal of respiratory and critical care medicine. 2018;198(5):e44-e68.

17. Suda T, Kaida Y, Nakamura Y, et al. Acute exacerbation of interstitial pneumonia associated with collagen vascular diseases. *Respiratory medicine.* 2009;103(6):846-853.

18. Park IN, Kim DS, Shim TS, et al. Acute exacerbation of interstitial pneumonia other than idiopathic pulmonary fibrosis. *Chest.* 2007;132(1):214-220.

19. Hozumi H, Nakamura Y, Johkoh T, et al. Acute exacerbation in rheumatoid arthritis-associated interstitial lung disease: a retrospective case control study. *BMJ open.* 2013;3(9):e003132.

20. Jennette JC, Falk RJ, Andrassy K, et al. Nomenclature of systemic vasculitides. Proposal of an international consensus conference. *Arthritis and rheumatism.* 1994;37(2):187-192.

21. Hozumi H, Oyama Y, Yasui H, et al. Clinical significance of myeloperoxidase-anti-neutrophil cytoplasmic antibody in idiopathic interstitial pneumonias. *PloS one.* 2018;13(6):e0199659.

22. Liu GY, Ventura IB, Achtar-Zadeh N, et al. Prevalence and Clinical Significance of Antineutrophil Cytoplasmic Antibodies in North American Patients With Idiopathic Pulmonary Fibrosis. *Chest.* 2019;156(4):715-723.

23. Kida T, Tanaka T, Yokota I, et al. Association between preexisting lung involvements and the risk of diffuse alveolar hemorrhage in patients with microscopic polyangiitis: A multi-center retrospective cohort study. *Modern rheumatology.* 2020;30(2):338-344.

24. Casian A, Jayne D. Management of alveolar hemorrhage in lung vasculitides. *Seminars in respiratory and critical care medicine.* 2011;32(3):335-345.

25. Guillevin L, Lhote F, Gayraud M, et al. Prognostic factors in polyarteritis nodosa and Churg-Strauss syndrome. A prospective study in 342 patients. *Medicine.*

1996;75(1):17-28.

26. Furuta S, Chaudhry AN, Hamano Y, et al. Comparison of phenotype and outcome in microscopic polyangiitis between Europe and Japan. *The Journal of rheumatology.* 2014;41(2):325-333.

27. Suzuki A, Sakamoto S, Kurosaki A, et al. Chest High-Resolution CT Findings of Microscopic Polyangiitis: A Japanese First Nationwide Prospective Cohort Study. *AJR. American journal of roentgenology.* 2019:1-11.

28. Shi J, Shen Q, Chen XM, Du XG. Clinical characteristics and outcomes in microscopic polyangiitis patients with renal involvement: a study of 124 Chinese patients. *BMC nephrology.* 2019;20(1):339.

29. Corral-Gudino L, Borao-Cengotita-Bengoa M, Del Pino-Montes J, Lerma-Márquez JL. Overall survival, renal survival and relapse in patients with microscopic polyangiitis: a systematic review of current evidence. *Rheumatology (Oxford, England).* 2011;50(8):1414-1423.

30. Alba MA, Flores-Suarez LF, Henderson AG, et al. Interstital lung disease in ANCA vasculitis. *Autoimmunity reviews.* 2017;16(7):722-729.

31. Jardel S, Puéchal X, Le Quellec A, et al. Mortality in systemic necrotizing vasculitides: A retrospective analysis of the French Vasculitis Study Group registry. *Autoimmunity reviews.* 2018;17(7):653-659.

32. Noble PW, Albera C, Bradford WZ, et al. Pirfenidone in patients with idiopathic pulmonary fibrosis (CAPACITY): two randomised trials. *Lancet (London, England).* 2011;377(9779):1760-1769.

33. Richeldi L, du Bois RM, Raghu G, et al. Efficacy and safety of nintedanib in idiopathic pulmonary fibrosis. *The New England journal of medicine.* 2014;370(22):2071-2082.

34. Flaherty KR, Wells AU, Cottin V, et al. Nintedanib in Progressive Fibrosing Interstitial Lung Diseases. *The New England journal of medicine.* 2019;381(18):1718-1727.

35. Distler O, Highland KB, Gahlemann M, et al. Nintedanib for Systemic Sclerosis-Associated Interstitial Lung Disease. *The New England journal of medicine.* 2019;380(26):2518-2528.

36. Maher TM, Corte TJ, Fischer A, et al. Pirfenidone in patients with unclassifiable progressive fibrosing interstitial lung disease: a double-blind, randomised, placebo-controlled, phase 2 trial. *The Lancet. Respiratory medicine.* 2020;8(2):147-157.

37. Ley B, Ryerson CJ, Vittinghoff E, et al. A multidimensional index and staging system for idiopathic pulmonary fibrosis. *Annals of internal medicine.* 2012;156(10):684-691.

38. Winstone TA, Assayag D, Wilcox PG, et al. Predictors of mortality and progression in scleroderma-associated interstitial lung disease: a systematic review. *Chest.* 2014;146(2):422-436.

39. Solomon JJ, Chung JH, Cosgrove GP, et al. Predictors of mortality in rheumatoid arthritis-associated interstitial lung disease. *The European respiratory journal.* 2016;47(2):588-596.

40. Fujisawa T, Hozumi H, Kono M, et al. Prognostic factors for myositis-associated interstitial lung disease. *PloS one.* 2014;9(6):e98824.

41. Kamiya Y, Fujisawa T, Kono M, et al. Prognostic factors for primary Sjogren's syndrome-associated interstitial lung diseases. *Respiratory medicine.* 2019;159:105811.

42. Kolb M, Bondue B, Pesci A, et al. Acute exacerbations of progressive-fibrosing

534 interstitial lung diseases. *European respiratory review : an official journal of the*
535 *European Respiratory Society.* 2018;27(150).
536 43. Leuschner G, Behr J. Acute Exacerbation in Interstitial Lung Disease. *Frontiers in*
537 *medicine.* 2017;4:176.
538 44. Birnbaum J, Danoff S, Askin FB, Stone JH. Microscopic polyangiitis presenting as a
539 "pulmonary-muscle" syndrome: is subclinical alveolar hemorrhage the mechanism of
540 pulmonary fibrosis? *Arthritis and rheumatism.* 2007;56(6):2065-2071.
541

542 **Table 1. Patient characteristics of MPA-ILD and MPA-non-ILD**

	MPA-ILD N = 84	**MPA-non-ILD N = 95**	***P*-value**
Age, years	73.8 ± 8.7	72.6 ± 12.9	0.77
Sex, Man	56 (66.7)	34 (35.8)	<0.01
Former or current smoker	57 (67.9)	30 (31.6)	<0.01
Diagnosis, Clinical/pathological	32 (38.1)/ 52 (61.9)	32 (33.7)/ 63 (66.3)	0.64
MPO-ANCA, titer to cut-off ratio	65.6 ± 142.0	35.6 ± 30.3	0.06
C-reactive protein, mg/dL	7.72 ± 6.39	6.13 ± 6.19	0.09
Manifestation/involvement, yes †			
General symptom	31 (36.9)	52 (54.7)	0.02
Cutaneous	25 (29.8)	19 (20.0)	0.16
Mucous membranes/eyes	5 (6.0)	1 (1.1)	0.10
Ear/nose/throat	5 (6.0)	5 (5.3)	1.00
Cardiovascular	10 (11.9)	10 (10.5)	0.82
Abdominal	7 (8.3)	13 (13.7)	0.34
Renal	73 (86.9)	92 (96.8)	0.02
Nerves system	19 (22.6)	15 (15.8)	0.26
Five-factor score, 0/ 1/ 2	6 (7.1)/50 (59.5)/ 28 (33.3)	2 (2.1)/ 67 (70.5)/ 25 (26.3)	0.17
KL-6, U/mL	681 ± 689	–	–
PaO_2, Torr	76.4 ± 15.0	–	–
% FVC, %	84.8 ± 17.0	–	–
% DL_{CO}, % §	73.8 ± 21.4	–	–
HRCT pattern			
UIP	33 (39.3)	–	–
Probable UIP	14 (16.7)	–	–
Indeterminate for UIP	11 (13.1)	–	–
Alternative	26 (30.9)	–	–
ILD-preceding MPA by ≥ 3 months	42 (50)	–	–
Induction treatment			
Corticosteroids	84 (100)	95 (100)	–
with an immunosuppressant	31 (36.9)	23 (24.2)	0.07
Cyclophosphamide	26	16	
Azathioprine	5	6	
Methotrexate	0	1	
with plasmapheresis	1 (1.2)	2 (2.1)	1.00
Observation period, months	43.9 ± 40.1	57.1 ± 58.2	0.08

AE development after MPA onset ‡	16 (19.1)	–	
Death during observation period	51	26	
Cause of death	N = 51	N = 26	<0.01
Pulmonary involvement-related			
ILD	17 (33.3)	0 (0)	
AE	13	0	
Chronic respiratory failure	4	0	
Non-ILD	0 (0)	1 (3.8)	
Infection	10 (19.6)	10 (38.5)	
Bleeding-related	12 (23.5)	6 (23.1)	
DAH	7	1	
Cerebral hemorrhage	1	3	
Gastrointestinal bleeding	4	2	
Cardiovascular event	6 (11.8)	3 (11.5)	
Uremia	3 (5.9)	4 (15.4)	
Malignancy	3 (5.9)	2 (7.7)	

Data presented as mean ± standard deviation or frequency (%).

† Other than pulmonary involvement

§ N = 49.

‡ median AE-free period after MPA diagnosis was 18.6 months (interquartile range: 3.7–35.9 months).

MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; MPA, microscopic polyangiitis; ILD, interstitial lung disease; KL-6, Krebs von den Lungen-6; PaO_2, arterial oxygen pressure; %FVC, predicted forced vital capacity; $\%DL_{CO}$, percent-predicted diffusing capacity of the lung carbon monoxide; HRCT, high-resolution computed tomography; UIP, usual interstitial pneumonia; AE, acute exacerbation; DAH, diffuse alveolar hemorrhage.

553 **Table 2. Characteristics of patients with MPA-ILD and ILD-alone**

	Matched MPA-ILD N = 80	**Matched ILD-alone N = 80**	***P*-value**
Age, years	73.2 ± 8.3	72.7 ± 7.4	0.70
Sex, Man	56 (70.0)	52 (65.0)	0.61
HRCT pattern, UIP/non-UIP	32 (40)/ 48 (60)	31 (38.8)/49 (61.2)	1.00
Former or current smoker	57 (71.3)	51 (63.8)	0.40
C-reactive protein, mg/dL	7.88 ± 6.46	1.05 ± 2.48	<0.01
KL-6, U/mL	682 ± 701	1034 ± 925	<0.01
PaO_2, Torr	76.8 ± 15.0	76.4 ± 13.5	0.89
% FVC, %	84.8 ± 17.3	74.2 ± 20.7	<0.01
% DL_{CO}, % †	74.1 ± 21.6	65.8 ± 19.8	0.08
Immunosuppressive treatment §			
Corticosteroids	80 (100)	23 (28.8)	<0.01
with an immunosuppressant	30 (37.5)	5 (6.3)	<0.01
Cyclophosphamide	26	2	
Cyclosporin	0	2	
Azathioprine	4	1	
Anti-fibrotic treatment §	0 (0)	18 (22.5)	<0.01
Pirfenidone	0‡	13	
Nintedanib	0	5	
Observation period, months	44.8 ± 40.7	47.7 ± 40.0	0.65
AE development after MPA onset	15 (18.8)	15 (18.8)	1.00
Death during observation period	50 (62.5)	30 (37.5)	<0.01
Cause of death	N = 50	N = 30	<0.01
ILD-related	17 (34.0)	21 (70.0)	
AE	13	10	
Chronic respiratory failure	4	11	
Infection	9 (18.0)	4 (13.3)	
Bleeding-related	12 (24.0)	0 (0)	
DAH	7	0	
Cerebral hemorrhage	1	0	
Gastrointestinal bleeding	4	0	
Cardiovascular event	6 (12.0)	2 (6.7)	
Uremia	3 (6.0)	0 (0)	
Malignancy	3 (6.0)	3 (10.0)	

554 Data presented as mean ± standard deviation or frequency (%).

† MPA-ILD, N = 47; ILD-alone, N = 38

§ Induction treatment after MPA diagnosis in patients with MPA or initial treatment after IIP diagnosis in patients with ILD-alone

‡ Only one patient in the ILD-preceding group was treated with pirfenidone prior to the clinical onset of MPA.

MPA, microscopic polyangiitis; ILD, interstitial lung disease; HRCT, high-resolution computed tomography; UIP, usual interstitial pneumonia; KL-6, Krebs von den Lungen-6; PaO_2, arterial oxygen pressure; %FVC, predicted forced vital capacity; $\%DL_{CO}$, percent-predicted diffusing capacity of lung carbon monoxide; AE, acute exacerbation; DAH, diffuse alveolar hemorrhage.

Table 3. Results of the Cox proportional hazards regression analysis of all-cause mortality

Variable	HR	95% CI	*P*-value
Univariate analysis			
Age, years	1.04	1.01–1.08	0.01
Man (vs. Woman)	1.18	0.66–2.23	0.59
Smoking, current/former (vs. never)	1.29	0.72–2.44	0.40
MPO-ANCA, per titer/cut-off ratio	1.00	0.99–1.01	0.93
C-reactive protein, per 1 mg/dL	1.01	0.97–1.05	0.70
Manifestation/involvement, yes (vs. no) †			
General symptom	0.67	0.35–1.20	0.18
Cutaneous	0.75	0.38–1.40	0.38
Mucous membranes/eyes	0.58	0.14–1.61	0.35
Ear/nose/throat	0.56	0.10–1.83	0.39
Cardiovascular	0.64	0.22–1.48	0.32
Abdominal	0.82	0.25–2.03	0.70
Renal	0.99	0.45–2.59	0.97
Nerves system	0.74	0.34–1.46	0.40
1996 five-factor score ≥ 2 (vs. <2)	0.78	0.40–1.53	0.47
KL-6, per 100 U/mL	1.02	0.98–1.05	0.24
PaO_2, per 1 Torr	0.97	0.95–0.99	<0.01
%FVC, per 1 %	0.96	0.94–0.98	<0.01
UIP pattern on HRCT, yes (vs. no)	1.46	0.83–2.55	0.18
Induction treatment regimen			
CS plus an IS (vs. CS monotherapy)	0.55	0.30–1.02	0.07
Multivariable analysis §			
Age, years	1.03	0.98–1.09	0.20
PaO_2, per 1 Torr	0.98	0.95–1.01	0.18
%FVC, per 1%	0.96	0.94–0.98	<0.01

† Other than pulmonary involvement

§ Age, sex, treatment, and all variables that showed a significant association in univariate analysis were included in the multivariable analysis.

HR, hazard ratio; 95% CI, 95% confidence interval; MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; KL-6, Krebs von den Lungen-6; PaO_2, arterial oxygen pressure; %FVC, predicted forced vital capacity; UIP, usual interstitial pneumonia; HRCT, high-resolution computed tomography; CS, corticosteroids; IS, immunosuppressant

Figure legends

Figure 1. Flow chart of this study

A Flow chart illustrating the selection of patients with MPA-ILD and patients with MPA-non-ILD

B Flow chart illustrating the selection of patients with MPA-ILD and patients with ILD-alone who were propensity-score matched for age, sex, and HRCT pattern

MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; MPA, microscopic polyangiitis; HRCT, high-resolution computed tomography; AE, acute exacerbation of interstitial lung disease; DAH, diffuse alveolar hemorrhage; CTD, connective tissue disease; PR3, proteinase 3; IIP, idiopathic interstitial pneumonia; UIP, usual interstitial pneumonia

Figure 2. Kaplan–Meier curves

A Survival rates of the MPA-ILD and the MPA-non-ILD groups

The 5-year cumulative survival rates of the MPA-ILD and the MPA-non-ILD groups were 46.1% and 74.3%, respectively ($P < 0.001$ by the log-rank test).

B Survival rates in the propensity score-matched MPA-ILD and ILD-alone groups

The 5-year cumulative survival rate in the propensity score matched MPA-ILD and ILD-alone groups were 46.4% and 58.0%, respectively ($P = 0.02$ by the log-rank test).

C AE incidence rates of the matched MPA-ILD and the matched ILD-alone groups

The 1-year cumulative incidence rates of the matched MPA-ILD group and the matched ILD-alone group were 7.5% and 5.2%, respectively ($P = 0.75$ by the Gray's test).

MPA, microscopic polyangiitis; ILD, interstitial lung disease; AE, acute exacerbation

600 **Figure 3.** AE and survival rates

601 The median survival time from AE onset was 0.76 months (interquartile range: 0.23–2.40),

602 and the post-AE 3-month survival rate was 25%.

603

604 AE, acute exacerbation

Take-Home Point

Question: What are the clinical features and prognostic factors in patients with microscopic polyangiitis (MPA) and interstitial lung disease (ILD)? What is the incidence of acute exacerbation (AE) of ILD in those patients, and what are their risk factors?

Results: Patients with MPA-ILD had a higher frequency of men and smokers and was associated with higher mortality than those with MPA without ILD. In patients with MPA-ILD, the 1-year AE cumulative incidence rate was 7.2%. A lower percent-predicted forced vital capacity was independently associated with a higher mortality rate and a higher AE incidence rate. Developing AE was strongly associated with shorter survival.

Interpretation: MPA-ILD represented a distinct phenotype with poor prognosis. The specific management for MPA-ILD and AE should be established.

Supplemental materials

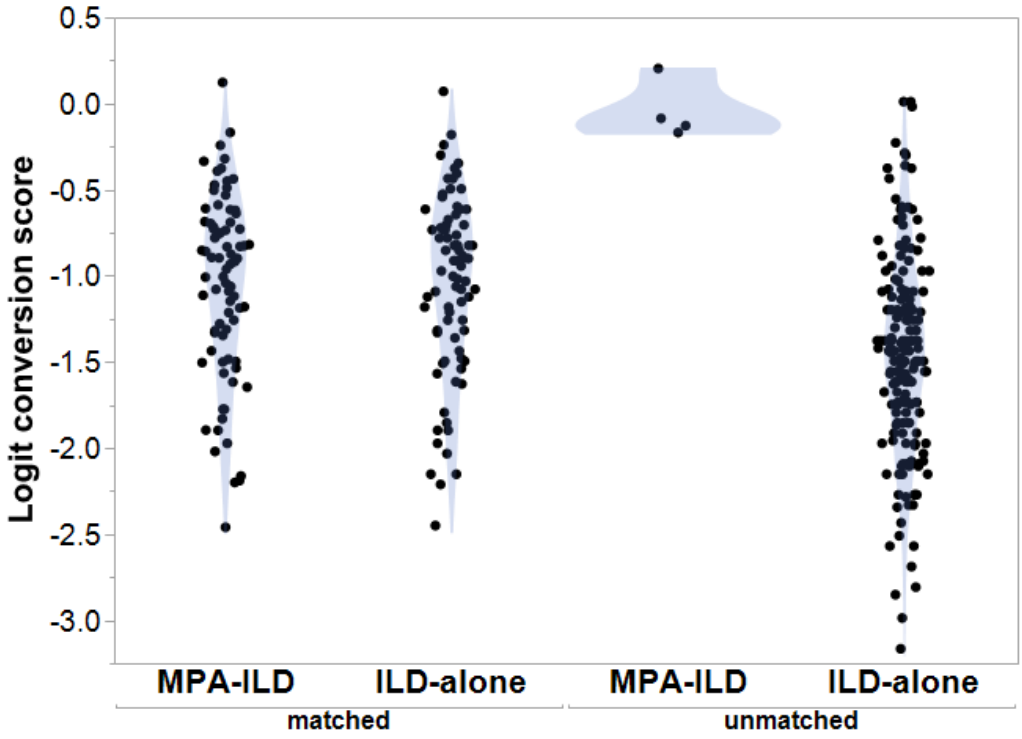
Title: Clinical significance of interstitial lung disease and its acute exacerbation in microscopic polyangiitis

Authors' full names:

Hironao Hozumi, M.D., Ph.D.[1]*, Masato Kono, M.D., Ph.D.[2], Hirotsugu Hasegawa, M.D., Ph.D.[3], Hideki Yasui, M.D., Ph.D.[1], Yuzo Suzuki, M.D., Ph.D.[1], Masato Karayama, M.D., Ph.D.[1], Kazuki Furuhashi, M.D., Ph.D.[1], Noriyuki Enomoto, M.D., Ph.D.[1]
Tomoyuki Fujisawa, M.D., Ph.D.[1], Naoki Inui, M.D., Ph.D.[1, 4], Yutaro Nakamura, M.D., Ph.D.[1], Koshi Yokomura, M.D., Ph.D.[2], Hidenori Nakamura, M.D., Ph.D.[3], Takafumi Suda, M.D., Ph.D.[1]

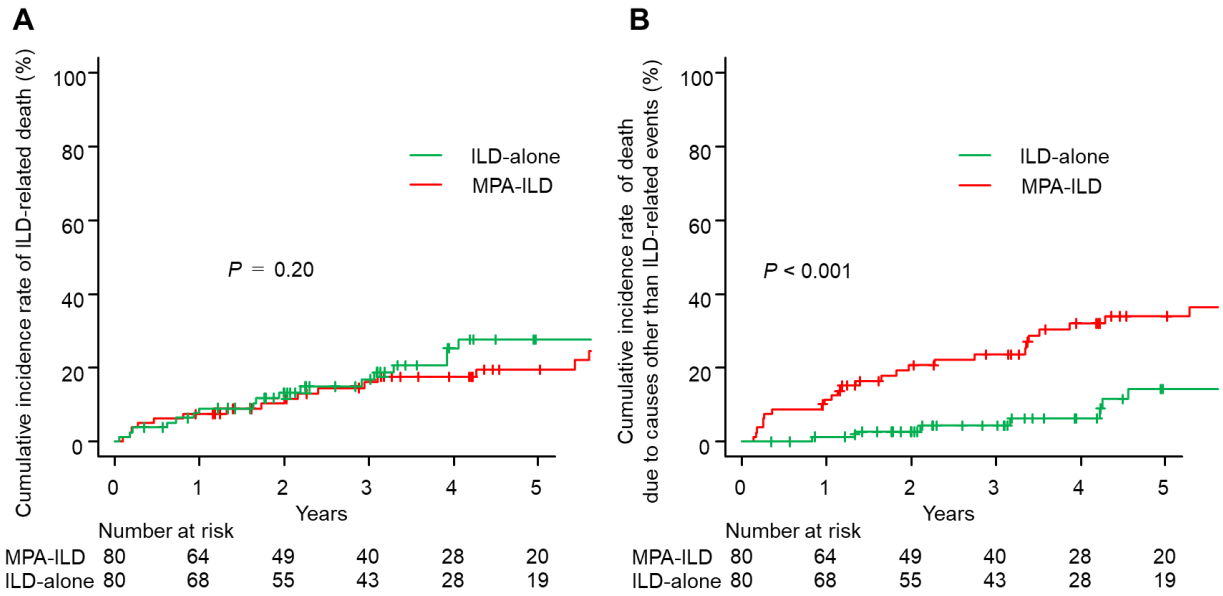
e-appendix 1.

Propensity scores, which are the predicted probabilities of each patient being assigned to MPA-ILD or ILD-alone, were calculated using a logistic regression model that was adjusted for sex (male or female) age and chest HRCT pattern (UIP or non-UIP) at the time of the diagnosis of MPA or IIP. Propensity score-matching was performed using the following algorithm: 1:1 nearest neighbour matching with a ± 0.05 caliper and no replacement. Based on this method, 80 age-, sex-, and chest HRCT pattern-matched pairs were made between the MPA-ILD and ILD-alone groups. Distribution of the logit conversion score in propensity score-matching is presented below.



MPA, microscopic polyangiitis; ILD, interstitial lung disease; IIP, idiopathic interstitial pneumonia; ILD-alone, IIP without the development of MPA or connective tissue disease and negative for myeloperoxidase-anti-neutrophil cytoplasmic antibody; HRCT, high-resolution computed tomography; UIP, usual interstitial pneumonia; MPO-ANCA, myeloperoxidase-anti-neutrophil cytoplasmic antibody

e-Figure 1. Incidence rates of death due to ILD-related or other causes



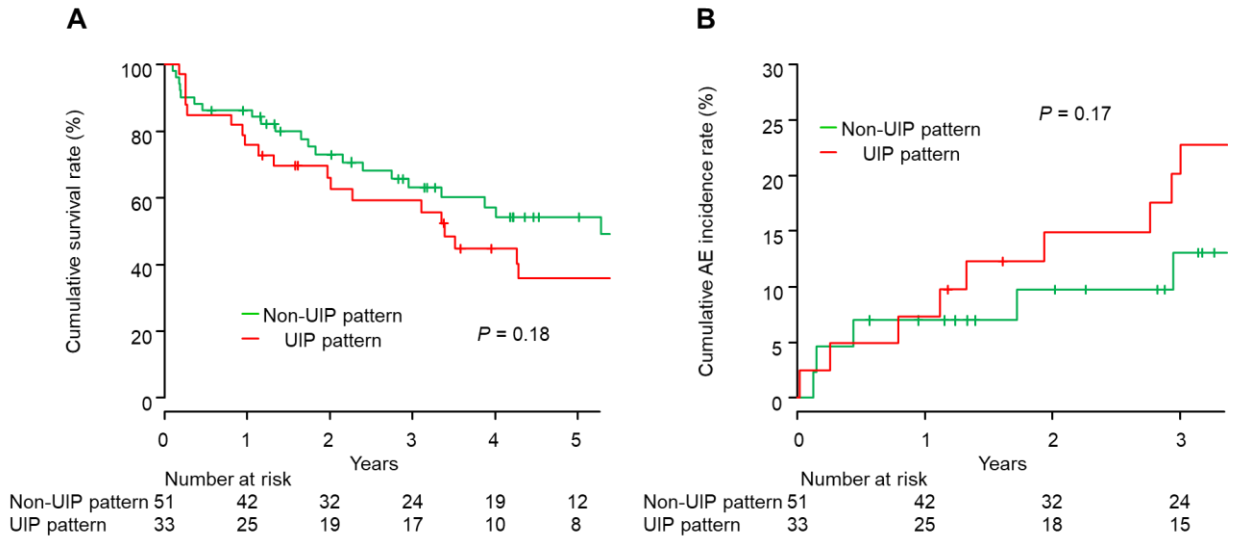
A Incidence rates of ILD-related death in the propensity score matched MPA-ILD and ILD-alone groups

The 5-year cumulative incidence rates in the propensity score matched MPA-ILD group and ILD-alone groups were 19.6% and 27.8%, respectively ($P = 0.20$ by the Gray's test).

B Incidence rates of death due to causes other than ILD-related related events in the propensity score matched MPA-ILD and ILD-alone groups

The 5-year cumulative incidence rates of the propensity score matched MPA-ILD and ILD-alone groups were 34.0% and 14.2%, respectively ($P < 0.001$ using the Gray's test).

e-Figure 2. Kaplan–Meier curves of patients disaggregated by HRCT patterns



A Survival rates by HRCT patterns

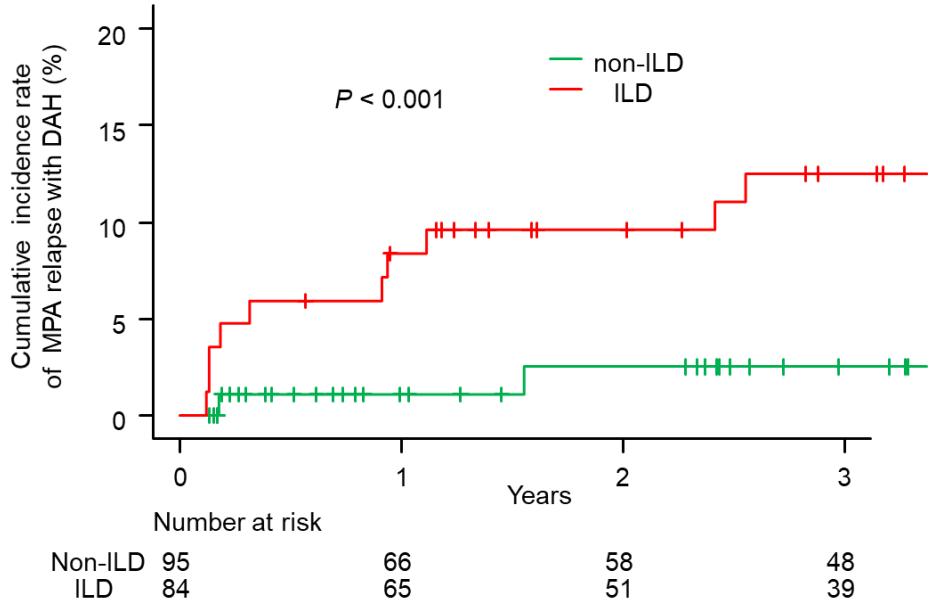
The 5-year cumulative survival rates in the HRCT-UIP pattern group and the HRCT-non-UIP pattern group were 35.8% and 54.2%, respectively ($P = 0.18$ by the log-rank test).

B AE incidence rates by HRCT patterns

The 1-year and 3-year cumulative incidence rates of the HRCT-UIP pattern group were 7.3% and 20.2%, respectively, while the 1-year and 3-year cumulative incidence rates of the HRCT-non-UIP pattern group were 7.0% and 13.1%, respectively ($P = 0.17$ using the Gray's test).

HRCT, high-resolution computed tomography; UIP, usual interstitial pneumonia; AE, acute exacerbation

e-Figure 3. Kaplan–Meier curves; sub-analysis on DAH



Incidence rates of MPA relapse with DAH in the MPA-ILD and the MPA-non-ILD groups
The 1-year and 3-year cumulative incidence rates in the MPA-ILD group were 8.4% and 12.5%, respectively, while the 1-year and 3-year cumulative incidence rates of the MPA-non-ILD group were 1.1% and 2.5%, respectively ($P < 0.001$ using the Gray's test).
DAH, diffuse alveolar hemorrhage; MPA, microscopic polyangiitis; ILD, interstitial lung disease

e-Table 1. Characteristics and pathological findings of ILD-preceding patients who underwent surgical lung biopsy

Age, years		Sex	MPO-ANCA†	HRCT pattern†	Pathological findings of the lung †				
ILD onset	MPA onset				Pattern	Vasculitis	Bronchiolitis	Lymphoid aggregates	Interstitial inflammation
59	70	Woman	unknown	Alternative	NSIP	–	+	+	+
61	71	Man	Negative	Alternative	NSIP	–	–	–	+
69	74	Woman	Negative	Alternative	NSIP	–	–	+	+
74	81	Man	unknown	Alternative	Unclassifiable	–	–	+	+
62	64	Man	Negative	UIP	UIP	–	–	+	+
52	68	Man	Positive	UIP	UIP	–	–	+	+
66	75	Man	Negative	UIP	UIP	–	+	–	+
65	74	Man	unknown	UIP	Unclassifiable	–	+	–	–

† at ILD diagnosis

\+ Present, – Absent

ILD, interstitial lung disease; MPA, microscopic polyangiitis; MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; HRCT, high-resolution computed tomography; UIP, usual interstitial pneumonia; NSIP, nonspecific interstitial pneumonia

e-Table 2. Results of the Fine-Gray subdistribution hazards analysis of ILD-related deaths

Variable	HR	95% CI	*P*-value
Univariate analysis			
Age, years	0.99	0.94–1.04	0.58
Man (vs. woman)	0.84	0.31–2.26	0.73
Smoking, current/former (vs. never)	1.12	0.39–3.24	0.83
MPO-ANCA, per titer/cut-off ratio	1.00	0.99–1.003	0.77
C-reactive protein, per 1 mg/dL	1.03	0.95–1.12	0.49
Manifestation/involvement, yes (vs. no) †			
General symptom	0.69	0.25–1.96	0.49
Cutaneous	0.48	0.14–1.70	0.26
Mucous membranes/eyes	0.95	0.22–4.07	0.94
Ear/nose/throat	0.74	0.10–5.40	0.77
Cardiovascular	1.03	0.22–4.93	0.97
Abdominal	0.65	0.07–5.65	0.69
Renal	1.02	0.22–4.78	0.98
Nerves system	1.56	0.67–3.67	0.30
1996 five-factor score ≥ 2 (vs. <2)	0.38	0.08–1.80	0.22
KL-6, per 100 U/mL	1.07	1.04–1.09	<0.01
PaO_2, per 1 Torr	0.98	0.95–1.01	0.26
%FVC, per 1 %	0.96	0.93–0.99	0.01
UIP pattern on HRCT, yes (vs. no)	1.58	0.63–3.96	0.33
Induction treatment with CS plus an IS (vs. CS monotherapy)	1.50	0.59–3.80	0.39
Multivariable analysis model 1 §			
KL-6, per 100 U/mL	1.07	1.03–1.12	<0.01
%FVC, per 1%	0.98	0.94–1.02	0.32
Multivariable analysis model 2 ‡			
KL-6, per 100 U/mL	1.07	1.03–1.10	<0.01
Multivariable analysis model 3 ‡			
%FVC, per 1%	0.96	0.92–0.99	0.01

† Other than pulmonary involvement

§ Age, sex, treatment, and all other variables that showed a significant association in univariate analysis were included in the multivariable analysis.

‡ Adjusted for age, sex, and treatment

HR, hazard ratio; 95% CI, 95% confidence interval; MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; KL-6, Krebs von den Lungen-6; PaO_2,

arterial oxygen pressure; %FVC, predicted forced vital capacity; UIP, usual interstitial pneumonia; HRCT, high-resolution computed tomography; CS, corticosteroids; IS, immunosuppressant

e-Table 3. Bronchoalveolar lavage fluid findings at the time of development of AE

	N = 16
Total cell count, × 10^5 cells/mL	1.3 (0.99–2.75)
Alveolar macrophages, %	77.7 (61.6–81.8)
Lymphocytes, %	9.5 (5.1–15.2)
Neutrophils, %	6.2 (1.9–17.4)
Eosinophils, %	2.0 (0.5–6.2)
CD4 / CD8, ratio	1.56 (0.84–2.15)

Data presented as median (interquartile range).

AE, acute exacerbation; CD, cluster of differentiation.

e-Table 4. Results of the Fine-Gray sub-distribution hazards analysis of AE development

Variable	HR	95% CI	*P*-value
Univariate analysis			
Age, years	1.00	0.94–1.06	0.87
Man (vs. woman)	1.44	0.46–4.57	0.53
Smoking, current/former (vs. never)	1.41	0.44–4.50	0.56
MPO-ANCA, per titer/cut-off ratio	1.00	0.99–1.01	0.66
C-reactive protein, per 1 mg/dL	1.03	0.94–1.12	0.54
Manifestation/involvement, yes (vs. no) †			
General symptom	0.54	0.17–1.71	0.30
Cutaneous	0.78	0.25–2.38	0.66
Mucous membranes/eyes	0.32	0.04–2.57	0.29
Ear/nose/throat	1.07	0.16–7.04	0.94
Cardiovascular	1.16	0.25–5.47	0.85
Abdominal	0.71	0.08–6.18	0.76
Renal	0.57	0.17–1.96	0.37
Nerves system	1.57	0.67–3.66	0.30
1996 five-factor score ≥ 2 (vs. <2)	0.44	0.10–2.04	0.29
KL-6, per 100 U/mL	1.02	0.98–1.07	0.35
PaO_2, per 1 Torr	1.00	0.96–1.04	0.88
%FVC, per 1 %	0.96	0.93–0.99	0.01
UIP pattern on HRCT (vs. non-UIP pattern)	2.47	0.91–6.70	0.07
Induction treatment with CS plus an IS (vs. CS monotherapy)	0.78	0.27–2.23	0.64
Multivariable analysis model §			
%FVC, per 1 %	0.96	0.93–0.99	0.01

† Other than pulmonary involvement

§ Age, sex, treatment, and all variables that showed a significant association in univariate analysis were included in the multivariable analysis.

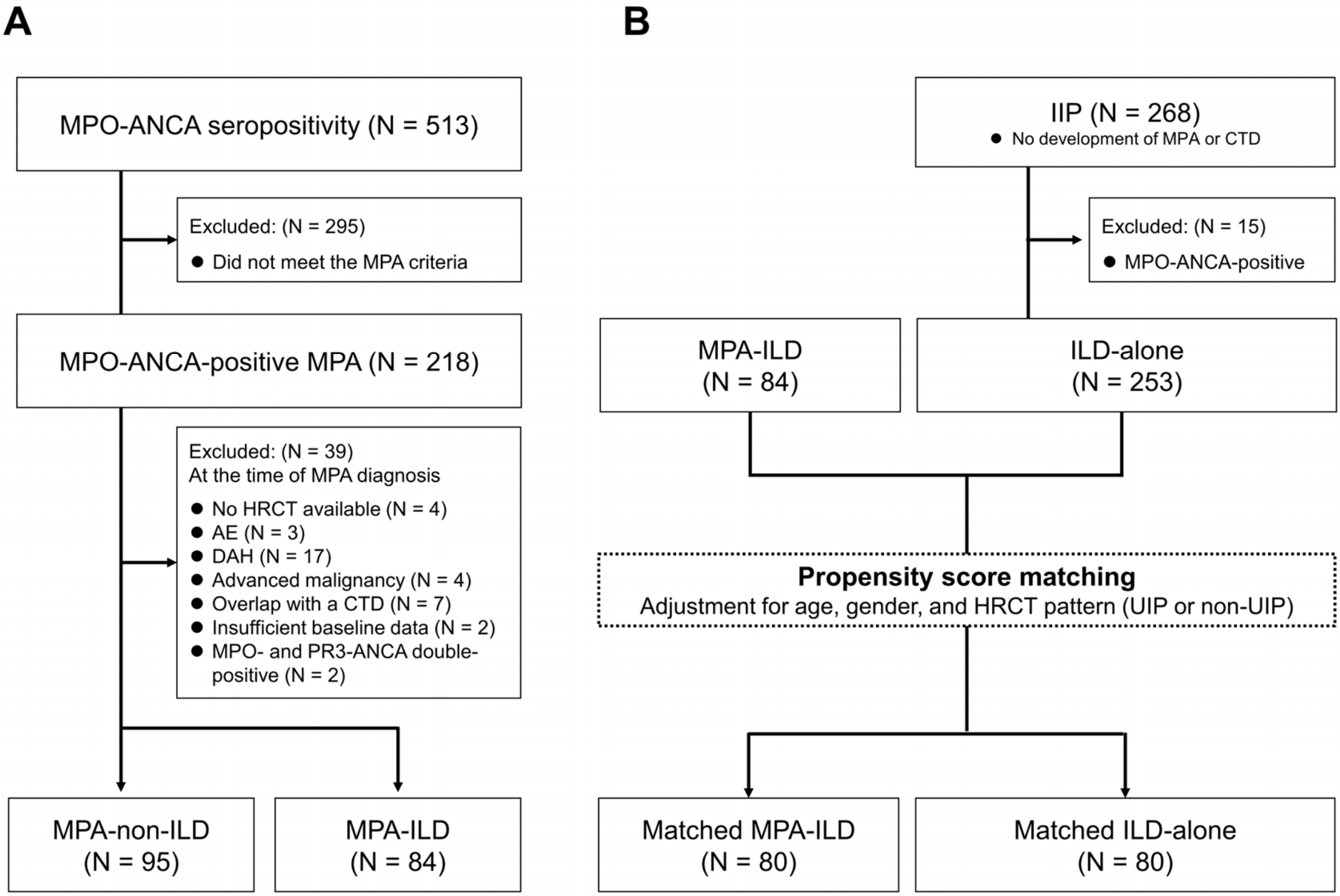
HR, hazard ratio; 95% CI, 95% confidence interval; MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; KL-6, Krebs von den Lungen-6; PaO_2, arterial oxygen pressure; %FVC, predicted forced vital capacity; UIP, usual interstitial pneumonia; HRCT, high-resolution computed tomography; CS, corticosteroids; IS, immunosuppressant

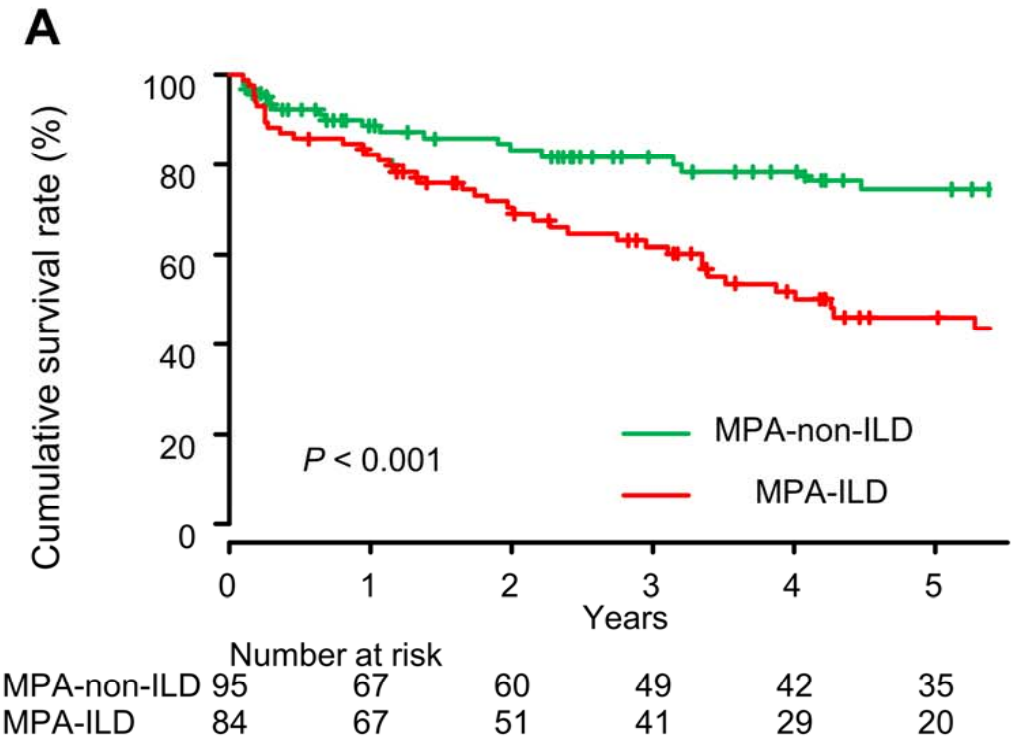
e-Table 5. Results of the Fine-Gray sub-distribution hazards analysis of MPA relapse with DAH

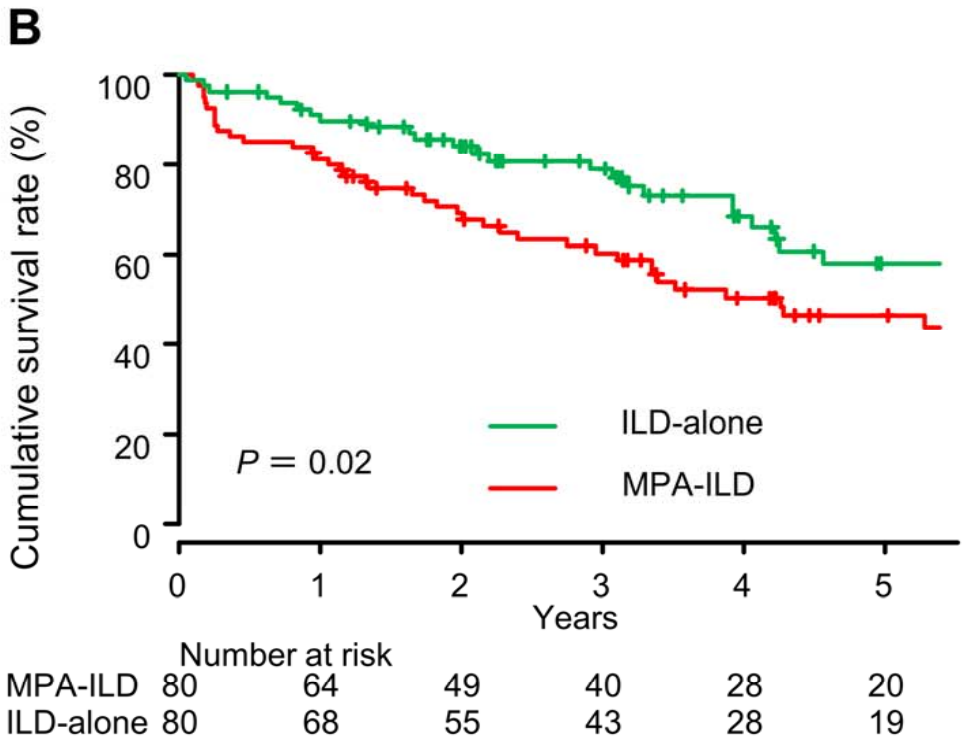
Variable	HR	95% CI	*P*-value
Univariate analysis			
Age, years	0.99	0.93–1.05	0.73
Man (vs. woman)	1.50	0.41–5.51	0.54
Smoking, current/former (vs. never)	2.51	0.55–11.5	0.23
MPO-ANCA, per titer/cut-off ratio	0.99	0.97–1.01	0.22
C-reactive protein, per 1 mg/dL	0.95	0.87–1.04	0.30
Manifestation/involvement, yes (vs. no) †			
General symptom	1.25	0.41–3.83	0.70
Cutaneous	0.80	0.22–2.98	0.74
Mucous membranes/eyes	3.09	0.74–12.9	0.12
Ear/nose/throat	1.55	0.20–11.7	0.67
Cardiovascular	1.53	0.35–6.80	0.58
Abdominal	0.77	0.25–2.35	0.64
Renal	0.71	0.16–3.16	0.66
Nerves system	1.20	0.34–4.27	0.78
1996 five-factor score ≥ 2 (vs. <2)	1.09	0.30–3.93	0.90
KL-6, per 100 U/mL	1.00	0.94–1.07	0.90
PaO_2, per 1 Torr	0.98	0.95–1.02	0.27
%FVC, per 1 %	0.99	0.97–1.02	0.49
UIP pattern on HRCT (vs. non-UIP pattern)	1.03	0.34–3.13	0.96
Induction treatment regimen			
CS plus an IS (vs. CS monotherapy)	0.82	0.26–2.66	0.75

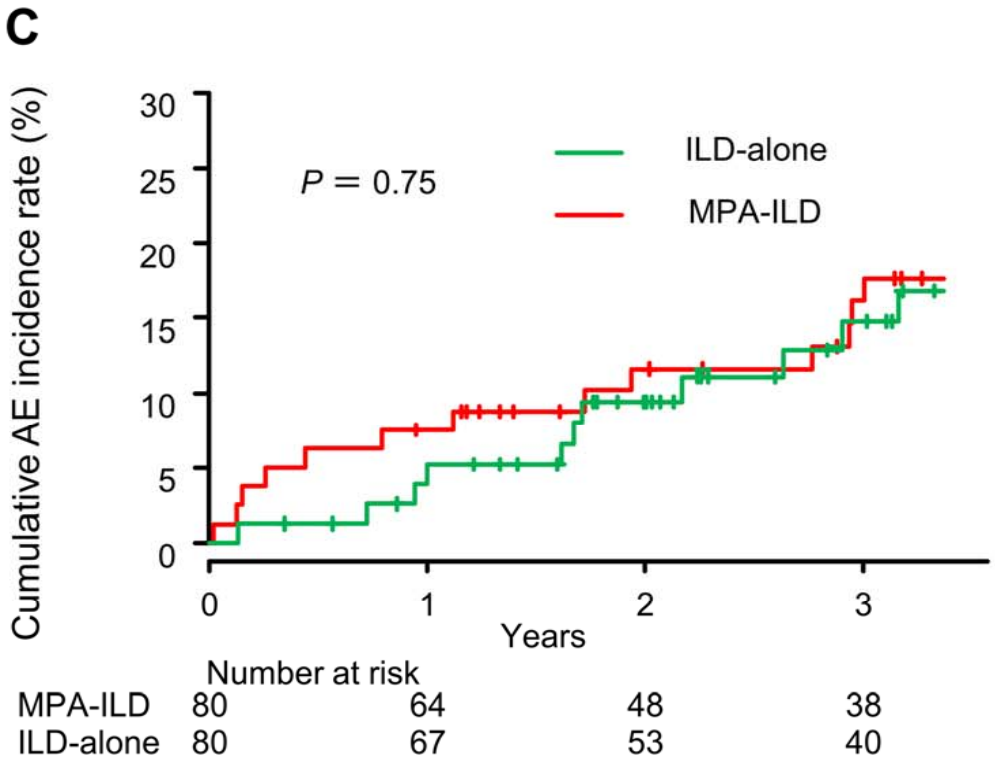
† Other than pulmonary involvement

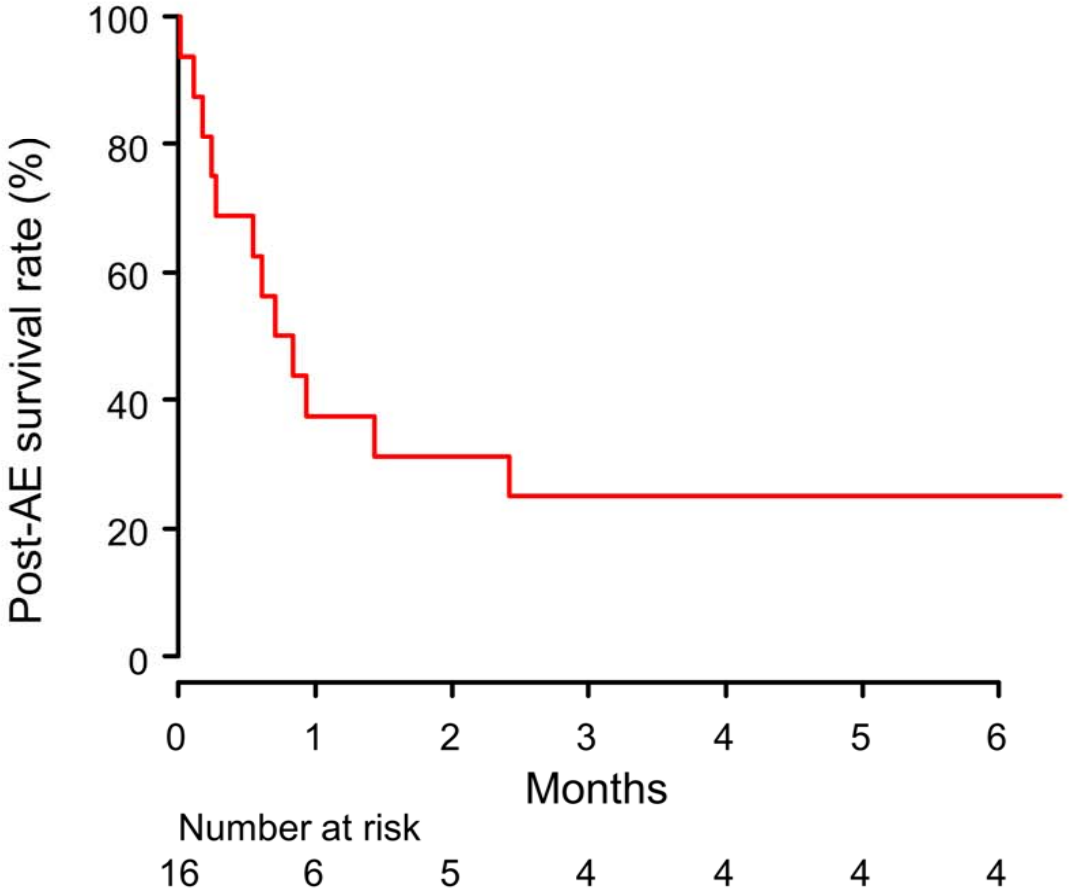
HR, hazard ratio; 95% CI, 95% confidence interval; MPO-ANCA, myeloperoxidase-antineutrophil cytoplasmic antibody; KL-6, Krebs von den Lungen-6; PaO_2, arterial oxygen pressure; %FVC, predicted forced vital capacity; UIP, usual interstitial pneumonia; HRCT, high-resolution computed tomography; CS, corticosteroids; IS, immunosuppressant


A
MPO-ANCA seropositivity (N = 513)
Excluded: (N = 295)
Did not meet the MPA criteria
MPO-ANCA-positive MPA (N = 218)
Excluded: (N = 39)
At the time of MPA diagnosis
No HRCT available (N = 4)
AE (N = 3)
DAH (N = 17)
Advanced malignancy (N = 4)
Overlap with a CTD (N = 7)
Insufficient baseline data (N = 2)
MPO- and PR3-ANCA double-positive (N = 2)
MPA-non-ILD (N = 95)
MPA-ILD (N = 84)
B
IIP (N = 268)
No development of MPA or CTD
Excluded: (N = 15)
MPO-ANCA-positive
MPA-ILD (N = 84)
ILD-alone (N = 253)
Propensity score matching
Adjustment for age, gender, and HRCT pattern (UIP or non-UIP)
Matched MPA-ILD (N = 80)
Matched ILD-alone (N = 80)


A
Cumulative survival rate (%)
100
80
60
40
20
0
P < 0.001
MPA-non-ILD
MPA-ILD
0
1
2
3
4
5
Years
Number at risk
MPA-non-ILD 95 67 60 49 42 35
MPA-ILD 84 67 51 41 29 20


B
Cumulative survival rate (%)
100
80
60
40
20
0
P = 0.02
ILD-alone
MPA-ILD
0
1
2
3
4
5
Years
Number at risk
MPA-ILD 80 64 49 40 28 20
ILD-alone 80 68 55 43 28 19


C
Cumulative AE incidence rate (%)
30
25
20
15
10
5
0
P = 0.75
ILD-alone
MPA-ILD
0
1
2
3
Years
Number at risk
MPA-ILD 80 64 48 38
ILD-alone 80 67 53 40


Post-AE survival rate (%)
100
80
60
40
20
0
0
1
2
3
4
5
6
Months
Number at risk
16
6
5
4
4
4
4