Monocyte Count as a Prognostic Biomarker in Patients with Idiopathic Pulmonary Fibrosis

Michael Kreuter1,2, Joyce S. Lee3, Argyrios Tzouvelekis4, Justin M. Oldham5, Philip L. Molyneaux6,7, Derek Weycker8, Mark Atwood8, Klaus-Uwe Kirchgaessler9, and Toby M. Maher6,7,10

1Center for Interstitial and Rare Lung Diseases, Pneumology, Thoraxklinik, University of Heidelberg, Heidelberg, Germany; 2German Center for Lung Research, Heidelberg, Germany; 3Department of Medicine, University of Colorado, Denver, Colorado; 4Department of Respiratory Medicine, University of Patras, Patras, Greece; 5Division of Pulmonary and Critical Care Medicine, Department of Internal Medicine, University of California Davis, Sacramento, California; 6Interstitial Lung Disease Unit, Royal Brompton Hospital, London, United Kingdom; 7Fibrosis Research Group, National Heart and Lung Institute, Imperial College London, London, United Kingdom; 8Policy Analysis, Inc., Brookline, Massachusetts; 9F. Hoffmann-La Roche, Ltd., Basel, Switzerland; and 10Hastings Center for Pulmonary Research and Division of Pulmonary, Critical Care, and Sleep Medicine, Keck School of Medicine, University of Southern California, Los Angeles, California.

ORCID IDs: 0000-0003-4957-8869 (J.M.O.); 0000-0001-7192-9149 (T.M.M.).

Abstract

Rationale: There is an urgent need for simple, cost-effective prognostic biomarkers for idiopathic pulmonary fibrosis (IPF); biomarkers that show potential include monocyte count.

Objectives: We used pooled data from pirfenidone and IFNγ-1b trials to explore the association between monocyte count and prognosis in patients with IPF.

Methods: This retrospective pooled analysis included patients (active and placebo arms) from the following four phase III, randomized, placebo-controlled trials: ASCEND (NCT01366209), CAPACITY (NCT00287729 and NCT00287716), and INSPIRE (NCT00075998). Outcomes included IPF progression (≥10% absolute decline in FVC% predicted, ≥50 mL decline in 6-minute walk distance, or death), all-cause hospitalization, and all-cause mortality over 1 year. The relationship between monocyte count (defined as time-dependent) and outcomes was assessed using bivariate and multivariable models.

Measurements and Main Results: This analysis included 2,067 patients stratified by monocyte count (at baseline: <6.00×10^9 cells/L [n = 1,609], 6.00 to <9.50×10^9 cells/L [n = 408], and ≥9.50×10^9 cells/L [n = 50]). In adjusted analyses, a higher proportion of patients with monocyte counts of 0.60 to <9.50×10^9 cells/L or ≥9.50×10^9 cells/L versus <6.00×10^9 cells/L experienced IPF progression (P = 0.016 and P = 0.002, respectively), all-cause hospitalization (P = 0.030 and P = 0.003, respectively), and all-cause mortality (P = 0.005 and P < 0.001, respectively) over 1 year. Change in monocyte count from baseline was not associated with any of the outcomes over 1 year and did not appear to be affected by study treatment.

Conclusions: In patients with IPF, elevated monocyte count was associated with increased risks of IPF progression, hospitalization, and mortality. Monocyte count may provide a simple and inexpensive prognostic biomarker in IPF.

Keywords: prognosis; pulmonary fibrosis; biomarkers
At a Glance Commentary

Scientific Knowledge on the Subject: The disease course of idiopathic pulmonary fibrosis (IPF) is highly variable, and prognosis can be difficult to predict, creating an urgent need for simple, cost-effective prognostic biomarkers to identify patients at risk of more rapid disease progression. Several potential prognostic biomarkers have been identified, but measurement of these biomarkers has been relatively complex, labor intensive, and costly. A recent retrospective analysis identified monocyte count as a potential prognostic biomarker for IPF, finding that high monocyte counts (>0.95 x 10^9 cells/L) were associated with an increased risk of mortality (vs. <0.95 x 10^9 cells/L).

What This Study Adds to the Field: In this retrospective pooled analysis of the ASCEND, CAPACITY, and INSPIRE trials of patients with IPF, elevated monocyte count was associated with significantly increased risks of IPF progression, hospitalization, and mortality over 1 year. In addition to a monocyte count of ≥0.95 x 10^9 cells/L, which was investigated in the previous study, our analysis also demonstrated that a monocyte count of 0.60 to <0.95 x 10^9 cells/L was associated with worse 1-year outcomes (vs. <0.60 x 10^9 cells/L). Our findings provide a rationale for prospective clinical studies investigating monocyte count as a simple and inexpensive prognostic biomarker for IPF.

Methods

Patient Population
This retrospective, pooled analysis included data from four phase III, randomized, placebo-controlled trials in IPF, as follows: ASCEND, CAPACITY (two studies), and INSPIRE. The eligibility criteria have been described previously (5, 6, 26) (see Appendix E1 in the online supplement). Patients who received 2,403 mg/d pirfenidone, 200 μg IFN-γ-1b three times a week, or placebo were included in this analysis (patients who received 1,197 mg/d pirfenidone in CAPACITY Study 004 were excluded) (5, 6, 26).

The trials were conducted in accordance with International Conference on Harmonization Guidelines and the Declaration of Helsinki. All patients provided written, informed consent before participation, and study protocols were approved by the institutional review board/ethics committee at each center.

Sample Collection and Measurements
Monocyte count was recorded at baseline and scheduled trial visits (see Appendix E1) as part of complete blood counts and was defined as a time-dependent variable in analyses described herein (i.e., monocyte count reflects the most recently available measurement on or before the time at which an event occurred). The thresholds used to stratify patients by monocyte count were defined initially in increments of 0.20 x 10^9 cells/L for values <0.80 x 10^9 cells/L (i.e., <0.20 x 10^9 cells/L, 0.20 to <0.40 x 10^9 cells/L, 0.40 to <0.60 x 10^9 cells/L, and 0.60 to <0.80 x 10^9 cells/L), whereas for values ≥0.80 x 10^9 cells/L, strata were defined as 0.80 to <0.95 x 10^9 cells/L and ≥0.95 x 10^9 cells/L on the basis of the analysis by Scott and colleagues that reported higher all-cause mortality among patients with monocyte count ≥0.95 x 10^9 cells/L (8). Strata for values <0.95 x 10^9 cells/L were subsequently modified on the basis of findings from bivariate analyses, suggesting that risks of study outcomes were comparable among patients within monocyte count strata defined as <0.60 x 10^9 cells/L (<0.20 x 10^9 cells/L, 0.20 to <0.40 x 10^9 cells/L, and 0.40 to <0.60 x 10^9 cells/L) and those defined as 0.60 to <0.95 x 10^9 cells/L (0.60 to <0.80 x 10^9 cells/L and 0.80 to <0.95 x 10^9 cells/L).

Results from a goodness-of-fit analysis corroborated the use of the modified thresholds for the analyses described herein. In supplemental analyses, other components of white blood cells (e.g., lymphocytes and neutrophils), which were recorded at baseline and scheduled trial visits (see Appendix E1), were also considered and were defined as continuous time-dependent variables.

Outcomes
Outcomes over 1 year were IPF progression (≥10% absolute decline in FVC% predicted,
Shared frailty models (an extension of the Cox proportional hazards model that adjusts for intracluster correlation [i.e., clustering by trial]) were employed to examine the relationship between monocyte count (defined as time-dependent), longitudinal change from baseline in monocyte count, and study outcomes over 1 year. Bivariate models were used without adjustment, and multivariable models were used with adjustment for patient demographics, physiologic function (time-dependent), comorbidity profile, and chronic immunosuppressant use (time-dependent). The monocyte stratification variable, change from baseline in monocyte count variable, and dummy variables for pirfenidone and IFN-1b were forced into models as regressors; other predictors of outcomes were selected for inclusion in the multivariable models via a backward selection method ($P < 0.10$).

Supplemental analyses of monocyte count and counts of other components of white blood cells were similarly conducted. Survival analyses were performed using Kaplan-Meier techniques, and monocyte count comparisons were undertaken using the log-rank test. For time-to-event analyses, patients were censored at the time of loss to follow-up or lung transplantation (or death) or censor.

### Statistical Analysis

Patients in the intent-to-treat populations with baseline monocyte count data were included in analyses; there was no imputation for missing values.

### Table 1. Summary of Patient Baseline Characteristics in the Pooled ASCEND, CAPACITY, and INSPIRE Population by Monocyte Count

| Monocyte Count | <0.60 × 10^9 cells/L | 0.60 to <0.95 × 10^9 cells/L | ≥0.95 × 10^9 cells/L |
|----------------|----------------------|-----------------------------|---------------------|
| Age, mean (SD), yr | 66.7 (7.6) | 66.4 (7.7) | 67.8 (7.3) | 67.7 (8.1) |
| Sex, n (%) | | | | |
| M | 1,508 (73.0) | 1,136 (70.6) | 328 (80.4) | 44 (88.0) |
| Monocytes, × 10^9 cells/L | | | | |
| Mean (SD) | 0.49 (0.18) | 0.41 (0.10) | 0.71 (0.09) | 1.14 (0.29) |
| Quintile, minimum–maximum | | | | |
| First | 0.10–0.34 | 0.10–0.32 | 0.60–0.62 | 0.95–0.96 |
| Second | 0.35–0.42 | 0.33–0.38 | 0.63–0.66 | 0.97–1.01 |
| Third | 0.43–0.50 | 0.39–0.44 | 0.67–0.71 | 1.02–1.09 |
| Fourth | 0.51–0.61 | 0.45–0.51 | 0.72–0.77 | 1.11–1.21 |
| Fifth | 0.62–2.81 | 0.52–0.59 | 0.78–0.94 | 1.24–2.81 |
| FVC% predicted, mean (SD) | 69.0 (15.0) | 68.7 (15.1) | 67.4 (14.3) | 63.1 (14.3) |
| DCO% predicted, mean (SD) | 43.6 (11.4) | 44.1 (11.4) | 41.9 (11.2) | 41.4 (11.4) |
| 6MWD, mean (SD), m | 382.0 (120.6) | 385.3 (120.5) | 374.7 (119.6) | 336.3 (122.8) |
| UCSD-SOBQ total score, mean (SD) | 39.1 (24.6) | 38.6 (24.4) | 39.7 (24.9) | 47.0 (26.8) |
| Comorbidities, n (%) | | | | |
| GERD | 645 (31.2) | 473 (29.4) | 154 (37.7) | 18 (36.0) |
| CAD | 444 (21.5) | 310 (19.3) | 111 (27.2) | 23 (46.0) |
| COPD | 114 (5.5) | 83 (5.2) | 30 (7.4) | 1 (2.0) |
| MI | 111 (5.4) | 73 (4.5) | 34 (8.3) | 4 (8.0) |
| Pulmonary hypertension | 50 (2.4) | 27 (1.7) | 22 (5.4) | 1 (2.0) |
| DVT | 48 (2.3) | 39 (2.4) | 8 (2.0) | 1 (2.0) |
| CV risk factors, n (%) | | | | |
| Smoker | 1,372 (66.4) | 1,064 (66.1) | 276 (67.6) | 32 (64.0) |
| Hypertension | 1,068 (51.7) | 815 (50.7) | 230 (56.4) | 23 (46.0) |
| Obesity | 897 (43.4) | 686 (42.6) | 187 (45.8) | 24 (48.0) |
| Hypercholesterolemia | 864 (41.8) | 650 (40.4) | 190 (46.6) | 24 (48.0) |
| Diabetes | 454 (22.0) | 348 (21.6) | 95 (23.3) | 11 (22.0) |
| Chronic immunosuppressant use, n (%) | | | | |
| Steroid (systemic) | 135 (6.5) | 102 (6.3) | 24 (5.9) | 9 (18.0) |
| Nonsteroid | 33 (1.6) | 25 (1.6) | 7 (1.7) | 1 (2.0) |
| Neither | 1,918 (92.8) | 1,486 (93.0) | 382 (93.6) | 40 (80.0) |

### Definition of abbreviations:
- 6MWD = 6-minute-walk distance
- CAD = coronary artery disease
- COPD = chronic obstructive pulmonary disease
- CV = cardiovascular
- DVT = deep vein thrombosis
- GERD = gastroesophageal reflux disease
- MI = myocardial infarction
- UCSD-SOBQ = University of California, San Diego, Shortness of Breath Questionnaire

$^a$At time of event (idiopathic pulmonary fibrosis progression, defined as >10% absolute decline in FVC% predicted, >50 m decline in 6MWD, or death) or censor.

$^b$Current/former.

$^c$Body mass index >30 kg/m².

$^d$At any time from baseline through to end of 1-year follow-up period; values may sum more than 100%.
mortality for all-cause hospitalization only) or at the end of 1-year follow-up, whichever occurred first.

**Results**

**Patient Baseline Characteristics**

Overall, 2,067 patients (ASCEND, n = 555; CAPACITY, n = 692; INSPIRE, n = 820) were included in this analysis. The patient demographics and disease characteristics have been previously described for the individual study populations (5, 6, 26) and are summarized for the pooled population in Table 1. The mean age (SD) for the pooled population was 66.7 (7.6) years, and 73.0% of patients were male (Table 1).

The mean (SD) monocyte count (at time of event [IPF progression] or censor) for the pooled population was 0.49 (0.18) × 10^9 cells/L (ASCEND, 0.52 [0.21] × 10^9 cells/L; CAPACITY, 0.48 [0.16] × 10^9 cells/L; INSPIRE, 0.47 [0.18] × 10^9 cells/L; Table E1). Other mean white blood cell counts at time of event or censor were also found to be relatively consistent across the included trials (Table E1). A total of 77.8% (n = 1,609), 19.7% (n = 408), and 2.4% (n = 50) of patients had baseline monocyte counts of <0.60 × 10^9 cells/L, 0.60 to <0.95 × 10^9 cells/L, and ≥0.95 × 10^9 cells/L, respectively. Physiologic characteristics at baseline were generally balanced between monocyte count groups, although patients in the ≥0.95 × 10^9 cells/L group had a numerically lower 6MWD and a numerically higher University of California, San Diego, Shortness of Breath Questionnaire score versus the <0.60 × 10^9 cells/L and 0.60 to <0.95 × 10^9 cells/L groups (Table 1).

The most frequently reported comorbidities in the overall population were gastroesophageal reflux disease (31.2%) and coronary artery disease (CAD; 21.5%; Table 1). CAD was more common in patients in the ≥0.95 × 10^9 cells/L monocyte group (46.0%) versus the <0.60 × 10^9 cells/L and 0.60 to <0.95 × 10^9 cells/L groups (19.3% and 27.2%, respectively). A higher percentage of patients in the ≥0.95 × 10^9 cells/L monocyte group (18.0%) had reported chronic systemic steroid immunosuppressant use versus the <0.60 × 10^9 cells/L and 0.60 to <0.95 × 10^9 cells/L groups (6.3% and 5.9%, respectively; Table 1).

**One-Year Risk of Study Outcomes by Monocyte Count**

Findings based on the initial stratification scheme for monocyte count are presented in Table E2. In bivariate analyses, after modification of the stratification scheme, times to first evidence of IPF progression, all-cause hospitalization, and all-cause mortality were shorter in patients with a monocyte count of ≥0.95 × 10^9 cells/L and 0.60 to <0.95 × 10^9 cells/L versus those with a count of <0.60 × 10^9 cells/L (Figure 1).

In adjusted analyses, a significantly higher percentage of patients with a monocyte count of either 0.60 to <0.95 × 10^9 cells/L or ≥0.95 × 10^9 cells/L versus those with a count of <0.60 × 10^9 cells/L experienced IPF progression (hazard ratio [HR], 1.25; 95% confidence interval [CI], 1.04–1.50; P = 0.016 and HR, 1.80; 95% CI, 1.23–2.63; P = 0.002, respectively), all-cause hospitalization (HR, 1.29; 95% CI, 1.03–1.63; P = 0.030 and HR, 2.14; 95% CI, 1.30–3.51; P = 0.003, respectively), and all-cause mortality (HR, 1.78; 95% CI, 1.19–2.66; P = 0.005 and HR, 4.05; 95% CI, 2.00–8.19; P < 0.001, respectively) over 1 year (Figure 2 and Table E3). This pattern was maintained when the models were adjusted only for age, sex, FVC% predicted, DLCO% predicted, and 6MWD (Table E4) and when nonchronic immunosuppressant use was considered (Table E5).

When other white blood cell counts were added to the adjusted multivariable analyses that included all model covariates, significant associations were observed between neutrophil count (positive association) and lymphocyte count (negative association) and study outcomes (Table E6). However, associations between elevated monocyte counts and increased risk of study outcomes were still observed when other white blood cell counts were accounted for in the adjusted model, which were significant for increased risk of IPF progression (for monocyte counts of 0.60 to <0.95 × 10^9 cells/L and ≥0.95 × 10^9 cells/L vs. <0.60 × 10^9 cells/L; HR, 1.22; 95% CI, 1.01–1.48; P = 0.039 and HR, 1.61; 95% CI, 1.08–2.39; P = 0.019, respectively) and all-cause hospitalization (for monocyte counts of ≥0.95 × 10^9 cells/L vs. <0.60 × 10^9 cells/L; HR, 1.75; 95% CI, 1.05–2.93; P = 0.031) over 1 year (Table E6).

**One-Year Risk of Study Outcomes by Change in Monocyte Count from Baseline**

The mean monocyte count ranged from 0.47 to 0.53 × 10^9 cells/L over the 1-year follow-up period (Figure E1), and large changes in monocyte count from baseline were rare (n = 18 or 19 for change of ≥0.50 × 10^9 cells/L). In the unadjusted model, changes in monocyte count of 0.10 to <0.50 × 10^9 cells/L (HR, 1.26; 95% CI, 1.01–1.58; P = 0.042) or ≥0.50 × 10^9 cells/L (HR, 2.46; 95% CI, 1.36–4.43; P = 0.003) from baseline were significantly associated with IPF progression versus a change of <−0.10 × 10^9 cells/L over 1 year (Table E7). Change in monocyte count of ≥0.50 × 10^9 cells/L from baseline was significantly associated with all-cause hospitalization versus a change of <−0.10 × 10^9 cells/L (HR, 2.31; 95% CI,
Study outcome n % with event Hazard ratios (95% CI) P value
IPF progression
Monocyte count, × 10^9 cells/L
<0.60 1,609 31.8 —
0.60–<0.95 408 40.0 0.016
>0.95 50 62.0 0.002
All-cause hospitalization
Monocyte count, × 10^9 cells/L
<0.60 1,614 18.3 —
0.60–<0.95 410 24.1 0.030
>0.95 43 39.5 0.003
All-cause mortality
Monocyte count, × 10^9 cells/L
<0.60 1,615 4.8 —
0.60–<0.95 405 8.9 0.005
>0.95 47 21.3 <0.001

Figure 2. Adjusted hazard ratios for IPF progression, all-cause hospitalization, and all-cause mortality by monocyte count. Monocyte count and other model covariates were defined as time-dependent variables, as appropriate. CI = confidence interval; IPF = idiopathic pulmonary fibrosis.

Discussion
This retrospective, pooled analysis found that patients with a confirmed diagnosis of IPF who had a monocyte count of 0.60 to <0.95 × 10^9 cells/L or ≥0.95 × 10^9 cells/L had a higher 1-year risk of IPF progression, all-cause hospitalization, and all-cause mortality versus patients with a monocyte count of <0.60 × 10^9 cells/L after adjustment for demographics, physiologic function, comorbidity profile, and chronic immunosuppressant use.

These findings are consistent with those of a previous study evaluating a possible link between monocyte count and prognosis in patients with IPF (8). In the previous retrospective, pooled analysis, a monocyte count of ≥0.95 × 10^9 cells/L was significantly associated with all-cause mortality versus a monocyte count of <0.95 × 10^9 cells/L after adjustment for FVC (HR, 2.47; P = 0.0063) and gender, age, and physiologic index (HR, 2.06; P = 0.0068) in a subset of patients with confirmed IPF. In the same study, a higher monocyte count was also associated with shortened survival in patients with other fibrotic diseases, including systemic sclerosis, myelofibrosis, and hypertrophic cardiomyopathy (8).

Further data supporting these findings come from an analysis of 231 patients from three Australian states in the Australian IPF registry (24). Analysis of these registry data found an association between monocyte counts of ≥0.95 × 10^9 cells/L and increased mortality after adjustment for age, sex, and baseline FVC% predicted (HR, 2.36; P = 0.02) (24).

Although the previous analyses only looked at ≥0.95 × 10^9 cells/L versus <0.95 × 10^9 cells/L (8, 24), we also demonstrated that a monocyte count of 0.60 to <0.95 × 10^9 cells/L was significantly associated with a higher 1-year risk of IPF progression, hospitalization, and mortality versus patients with a monocyte count of <0.60 × 10^9 cells/L. Moreover, this association was found after adjustment for age, sex, FVC% predicted, DLCO% predicted, and 6MWD, suggesting that monocyte count may provide added predictive value to these clinical variables. In contrast, we found that change from baseline in monocyte count over 1 year was not associated with study outcomes. This suggests that monocyte count has potential as a prognostic biomarker rather than as a predictive biomarker for treatment response, although this has not been confirmed yet in prospective cohorts.

Looking at real-world applications of our findings, a monocyte count of ≥0.60 × 10^9 cells/L could potentially be used to enrich the population of clinical trials for patients at greater risk of IPF progression. Furthermore, in clinical practice, a monocyte count of ≥0.60 × 10^9 cells/L.
could be used to alert healthcare professionals to a patient’s risk of IPF progression and worse prognosis, which may help to guide the decision to initiate treatment and the choice of therapeutic interventions. In addition, a monocyte count of $\geq 0.60 \times 10^9$ cells/L could be used to identify which patients to assess for their suitability for a lung transplant.

Preliminary analyses assessing the relationship between other white blood cell counts and study outcomes found significant associations for neutrophil count (positive association) and lymphocyte count (negative association) in multivariable analyses. However, elevated monocyte counts were still associated with a higher risk of study outcomes when the multivariable model was adjusted for other white blood cell counts, indicating that the relationship between monocyte counts and study outcomes is distinct from that of the other white blood cell counts. Additional analyses would be needed to further assess whether other white blood cell counts, apart from monocytes, have a relationship with the outcomes observed.

These results indicate an association between monocyte (and potentially other white blood cell) concentrations and IPF prognosis, which is in line with what is known about the possible roles of monocytes and other immune cells in the development of IPF and may help shed some light on the pathogenesis of IPF. Although the pathophysiology of IPF has not yet been fully elucidated, various immune cells have previously been linked with pathogenesis, including monocytes, neutrophils, and lymphocytes (27–33). The model of IPF pathogenesis currently favored is based on repeated epithelial injury leading, in genetically susceptible individuals, to aberrant repair and the formation of fibrotic tissue (34, 35). Fitting with this model, immune cells (including monocytes) migrate to the site of injury to aid repair, where they differentiate. Recruitment and differentiation of monocytes is driven by the surrounding microenvironment, with circulating monocytes having the potential to become interstitial or airway macrophages or dendritic cells (35–38). Disease progression in IPF has been linked to changes in the phenotype and function of alveolar macrophages. It follows that altered alveolar macrophage phenotype may be driven, in part, by changes in the populations of differentiated monocyte subsets, especially as monocyte-driven changes in airway macrophages are observed in healthy human aging (32, 39, 40). Single-cell RNA sequencing of lung collected from individuals with IPF has confirmed marked alveolar macrophage heterogeneity, something that has been linked to the evolution of fibrosis in murine models (41, 42). Accumulation of distinct populations of alveolar macrophages has been linked to disease progression and shortened survival in patients with IPF (43, 44).

The fibrocyte, a specialized cell derived from the monocyte cell lineage, has been postulated to be a precursor of the myofibroblast and has been implicated in the pathogenesis of IPF (45, 46). In patients with IPF, higher concentrations of circulating fibrocytes may be predictive of a worse prognosis in terms of disease progression and have shown to be markedly elevated in acute exacerbations of IPF (46, 47). C-C motif chemokine ligand 18, produced by fibrocytes and, to a greater extent, by alveolar macrophages, has shown potential as a serum biomarker of disease progression and mortality in IPF (45, 48, 49).

The emergence of readily measurable serum biomarkers that are reflective of IPF-related pathophysiology may help to better inform treatment approaches in IPF. In addition to the prediction of poorer prognosis, monocyte count has been linked with the occurrence of acute exacerbations of fibrosing ILDs, and regular monitoring may help to guide clinical decision-making with respect to the initiation of antifibrotic medications (8, 24, 50). The ease and speed with which biomarkers can be measured will govern their applicability within a clinical setting. Monitoring of circulating monocyte concentrations is easily incorporated into the routine assessment of patients with IPF as part of a standard blood count in clinical practice, and it represents a technically reproducible biomarker that is simpler, less labor intensive, and more cost-effective than measurement of other prognostic biomarkers identified for IPF, such as gene signatures (8, 15). Further prospective cohort studies are required to fully explore the relationships between monocyte count and change in monocytes from baseline on outcomes in IPF and response to treatment. These studies could also be designed to evaluate whether the findings in patients with IPF can be more broadly applied to patients with other forms of ILD.

There are a number of limitations that should be considered when interpreting the results of this analysis, including the relatively small number of patients included in the $\geq 0.95 \times 10^9$ cells/L monocyte count group versus the 0.60 to $<0.95 \times 10^9$ cells/L and $<0.60 \times 10^9$ cells/L groups. The low mortality rate across subgroups defined by change in monocyte count (and corresponding low statistical power) is another limitation. Furthermore, the effect of comorbidities (such as CAD, which may also result in high monocyte concentrations) (51) on the observed association between monocyte count and study outcomes is not known. In addition, steroid use can lead to reduced monocyte counts and possible shifts in monocyte phenotype (52), and although chronic steroid use was not found to considerably affect monocyte counts in this analysis, no data were available on the duration or timing of nonchronic steroid use, and, as such, their effect on monocyte counts could not be fully determined. We also considered that treatment group (placebo, pirfenidone, or IFN-γ-1b) may also have affected monocyte count; this possible effect could not be adequately assessed because classification and identification of patients who were responders to therapy was not possible in a robust manner with the current data set. In a descriptive analysis, although some differences in mean change from baseline in monocyte count over 1 year were observed for pirfenidone and IFN-γ-1b compared with placebo, these changes were not clinically meaningful. This was a retrospective, pooled analysis restricted to patients with IPF from clinical trials; therefore, the analysis population included more patients with greater short-term survival than real-world cohorts because patients with severe disease were excluded. This analysis may also underestimate the longer-term prognostic value of monocyte counts because relatively limited outcome data were available beyond the end of the trials. It should also be noted that the relationship between monocyte count and study outcomes did not appear to be linear, and thus a categorical (rather than continuous) measure for monocyte count was employed in most of these analyses.

Conclusions

In this retrospective analysis of pooled data from ASCEND, CAPACITY, and INSPIRE, elevated monocyte count was associated with
increased risks of IPF progression, hospitalization, and mortality. Monocyte count may provide a novel, simple, and inexpensive prognostic biomarker in patients with IPF and should be investigated further in future prospective clinical studies. ■

Author disclosures are available with the text of this article at www.atsjournals.org.

References

1. Ley B, Collard HR, King TE Jr. Clinical course and prediction of survival in idiopathic pulmonary fibrosis. Am J Respir Crit Care Med 2011;183:431–440.

2. Raghu G, Remy-Jardin M, Myers JL, Richeldi L, Ryerson CJ, Lederer DJ, et al.; American Thoracic Society, European Respiratory Society, Japanese Respiratory Society, and Latin American Thoracic Society. Diagnosis of idiopathic pulmonary fibrosis: an official ATS/ERS/JRS/ALAT clinical practice guideline. Am J Respir Crit Care Med 2018;198:e44–e68.

3. Nathan SD, Shlobin OA, Weir N, Ahmad S, Kalidj JM, Battle E, et al. Long-term course and prognosis of idiopathic pulmonary fibrosis in the new millennium. Chest 2011;140:221–229.

4. Vancheri C, Failla M, Crimi N, Raghu G. Idiopathic pulmonary fibrosis: a disease with similarities and links to cancer biology. Eur Respir J 2010;35:496–504.

5. King TE Jr, Bradford WZ, Castro-Bernardini S, Fagan EA, Glaspole I, et al.; ASCEND Study Group. A phase 3 trial of pirfenidone in patients with idiopathic pulmonary fibrosis. N Engl J Med 2014;370:2083–2092.

6. Noble PW, Albera C, Bradford WZ, Costabel U, Glassberg MK, Kardatzke D, et al.; CAPACITY Study Group. Pirfenidone in patients with idiopathic pulmonary fibrosis (CAPACITY): two randomised trials. Lancet 2011;377:1760–1769.

7. Richeldi L, du Bois RM, Raghu G, Azuma A, Brown KK, Costabel U, et al.; INPULSIS Trial Investigators. Efficacy and safety of nintedanib in idiopathic pulmonary fibrosis. N Engl J Med 2014;370:2071–2082.

8. Scott MKD, Quinn K, Li Q, Carroll R, Warsinske H, Vallania F, et al. Increased monocyte count as a cellular biomarker for poor outcomes in fibrotic diseases: a retrospective, multicentre cohort study. Lancet Respir Med 2019;7:497–508.

9. Neighbors M, Cabanski CR, Ramalingam TR, Sheng XR, Tew GW, Gu C, et al. Prognostic and predictive biomarkers for patients with idiopathic pulmonary fibrosis treated with pirfenidone: post-hoc assessment of the CAPACITY and ASCEND trials. Lancet Respir Med 2018;6:615–626.

10. Kohn N, Rossler A-K, Hornemann K, Muley T, Grünig E, Schmidt W, et al. C-proSP-B: a possible biomarker for pulmonary diseases? Respiration 2018;96:117–126.

11. Tzouvelekis A, Herazo-Mayado JD, Slade M, Chu JH, Deiuliis G, Pyu C, et al. Validation of the prognostic value of MMP-7 in idiopathic pulmonary fibrosis. Respiratory 2017;22:486–493.

12. Mahler TM, Oballa E, Simpson JK, Porte J, Habgood A, Fahy WA, et al. An epithelial biomarker signature for idiopathic pulmonary fibrosis: an analysis from the multicentre PROFILE cohort study. Lancet Respir Med 2017;5:946–955.

13. Jenkins RG, Simpson JK, Saini G, Bentley JH, Russell AM, Braybrooke R, et al. Longitudinal change in collagen degradation biomarkers in idiopathic pulmonary fibrosis: an analysis from the prospective, multicentre PROFILE study. Lancet Respir Med 2015;3:462–472.

14. Raghu G, Richeldi L, Jagerschmidt A, Martin V, Subramaniam A, Ozoux ML, et al. Idiopathic pulmonary fibrosis: prospective, case-controlled study of natural history and circulating biomarkers. Chest 2018;154:1359–1370.

15. Herazo-Mayado JD, Sun J, Molyneaux PL, Li Q, Villalba JA, Tzouvelekis A, et al. Validation of a 52-gene risk profile for outcome prediction in patients with idiopathic pulmonary fibrosis: an international, multicentre, cohort study. Lancet Respir Med 2017;5:857–868.

16. Korthagen NM, van Mooren CH, Barpo NP, Ruven HJ, Kruit A, Heron M, et al. Serum and BALF YKL-40 levels are predictors of survival in idiopathic pulmonary fibrosis. Respir Med 2011;105:106–113.

17. Stuurt BD, Lee JS, Kozlitina J, Noth I, Devine MS, Glazer CS, et al. Effect of telomere length on survival in patients with idiopathic pulmonary fibrosis: an observational cohort study with independent validation. Lancet Respir Med 2014;2:557–565.
36. Boyette LB, Macedo C, Hadi K, Elinoff BD, Walters JT, Ramaswami B, et al. Phenotype, function, and differentiation potential of human monocyte subsets. PLoS One 2017;12:e0176460.

37. Duffield JS, Lupher M, Thannickal VJ, Wynn TA. Host responses in tissue repair and fibrosis. Annu Rev Pathol 2013;8:241–276.

38. Byrne AJ, Maher TM, Lloyd CM. Pulmonary macrophages: a new therapeutic pathway in fibrosing lung disease? Trends Mol Med 2016;22:303–316.

39. Greiffo FR, Viteri-Alvarez V, Frankenberger M, Dietel D, Ortega-Gomez A, Lee JS, et al. CX3CR1-fractalkine axis drives kinetic changes of monocytes in fibrotic interstitial lung diseases. Eur Respir J 2020;55:pii1900460.

40. Byrne AJ, Powell JE, O’Sullivan BJ, Ogger PP, Hoffland A, Cook J, et al. Dynamics of human monocytes and airway macrophages during healthy aging and after transplant. J Exp Med 2020;217:e20191236.

41. Aran D, Looney AP, Liu L, Wu E, Fong V, Hsu A, et al. Reference-based analysis of lung single-cell sequencing reveals a transitional profibrotic macrophage. Nat Immunol 2019;20:163–172.

42. Adams TS, Schupp JC, Poli S, Ayaub EA, Neumark N, Ahangari F, et al. Single Cell RNA-seq reveals ectopic and aberrant lung resident cell populations in Idiopathic Pulmonary Fibrosis [preprint]. bioRxiv 2019. Available from: https://www.biorxiv.org/content/10.1101/759902v1.

43. Nouno T, Okamoto M, Ohnishi K, Kaieda S, Tominaga M, Zaizen Y, et al. Elevation of pulmonary CD163+ and CD204+ macrophages is associated with the clinical course of idiopathic pulmonary fibrosis patients. J Thorac Dis 2019;11:4005–4017.

44. Alden SJ, Ogger PP, Ghai P, McErlean P, Hewitt R, Toshner R, et al. The transferrin receptor CD71 delineates functionally distinct airway macrophage subsets during idiopathic pulmonary fibrosis. Am J Respir Crit Care Med 2019;200:209–219.

45. Walsh SM, Worrell JC, Fabre A, Hinz B, Kane R, Keane MP. Novel differences in gene expression and functional capabilities of myofibroblast populations in idiopathic pulmonary fibrosis. Am J Physiol Lung Cell Mol Physiol 2018;315:L697–L710.

46. Heukels P, van Hulst JAC, van Nimwegen M, Boorsma CE, Melgert BN, van den Toorn LM, et al. Fibrocytes are increased in lung and peripheral blood of patients with idiopathic pulmonary fibrosis. Respir Res 2018;19:90.

47. Moeller A, Gilpin SE, Ask K, Cox G, Cook D, Gauldie J, et al. Circulating fibrocytes are an indicator of poor prognosis in idiopathic pulmonary fibrosis. Am J Respir Crit Care Med 2009;179:588–594.

48. Prasse A, Probst C, Bargagli E, Zissel G, Toews GB, Flaherty KR, et al. Serum CC-chemokine ligand 18 concentration predicts outcome in idiopathic pulmonary fibrosis. Am J Respir Crit Care Med 2009;179:717–723.

49. Cai M, Bonella F, He X, Sitch SU, Sarria R, Guzman J, et al. CCL18 in serum, BAL fluid and alveolar macrophage culture supernatant in interstitial lung diseases. Respir Med 2013;107:1444–1452.

50. Kawamura K, Ichikado K, Anan K, Yasuda Y, Sekido Y, Sug M, et al. Monocyte count and the risk for acute exacerbation of fibrosing interstitial lung disease: a retrospective cohort study. Chron Respir Dis 2020;17:1479973120909840.

51. Ji H, Li Y, Fan Z, Zuo B, Jian X, Li L, et al. Monocyte/lymphocyte ratio predicts the severity of coronary artery disease: a syntax score assessment. BMC Cardiovasc Disord 2017;17:90.

52. Yeager MP, Pioi PA, Collins J, Barr F, Metzler S, Sites BD, et al. Glucocorticoids enhance the in vivo migratory response of human monocytes. Brain Behav Immun 2016;54:86–94.