Pulmonary involvement in long-term mixed connective tissue disease: functional trends and imaging findings after 10 years

L. Kawano-Dourado[1], B.G. Baldi[1], F.U. Kay[2], O.M. Dias[1], T.E.H. Gripp[2], P.S. Gomes[3], R. Fuller[4], M.T.C. Caleiro[4], R.A. Kairalla[1], C.R.R. Carvalho[1]

[1]Pulmonary Division, Heart Institute (InCor), and [2]Radiology Institute, University of Sao Paulo Medical School, Sao Paulo, Brazil;
[3]Pulmonary Division, Hospital do Servidor Publico Estadual (HSPE), Sao Paulo, Brazil;
[4]Rheumatology Division, Hospital das Clinicas, University of Sao Paulo Medical School, Sao Paulo, Brazil.

Abstract

Objective

Interstitial lung disease (ILD) is highly prevalent in patients with mixed connective tissue disease (MCTD). However, little is known about the long-term progression of ILD in MCTD. The aims of this study were to describe pulmonary function test (PFT) and high-resolution computed tomography (HRCT) results in long-term MCTD patients, to measure changes in PFT and HRCT results over a 10-year period, and to ascertain correlations in functional and imaging data.

Methods

In this retrospective cohort study, comparison between baseline and follow-up PFT and HRCT data was performed for 39 unselected consecutive MCTD patients.

Results

At baseline, 51% of the patients had abnormal PFTs. Forced vital capacity (FVC) was slightly reduced at baseline (77% of predicted), but remained stable after 10 years. A relative decrease of 15% in the diffusion capacity for carbon monoxide (DL_{CO}) was detected (from 84% to 71% of predicted, $p<0.001$). The median lower lobes ILD-HRCT score progressed from 7.5% at baseline to 11.2% at follow-up ($p=0.02$), and findings of traction bronchiolectasis and honeycombing increased ($p<0.05$). A moderate negative correlation was observed between functional parameters and quantification of image findings.

Conclusion

Functional and radiologic alterations suggestive of ILD in long-term MCTD patients are prevalent, mild, and progressed slightly over time. The most sensitive parameters for detecting subtle progression of ILD in MCTD patients are trends in DL_{CO}, quantification of lower-lobes disease by HRCT (lower-lobes %ILD-HRCT score), and qualitative analysis of HRCT imaging.

Key words

mixed connective tissue disease, interstitial lung disease, pulmonary function test, high-resolution computed tomography

Leticia Kawano-Dourado, MD
Bruno G. Baldi, MD
Fernando U. Kay, MD
Olivia M. Dias, MD
Thais EH. Gripp, MD
Paula S. Gomes, MD
Ricardo Fuller, MD
Maria T.C. Caleiro, MD
Ronaldo A. Kairalla, MD
Carlos R.R. Carvalho, MD

Please address correspondence to:
Leticia Kawano-Dourado, MD,
Av Dr Eneas de Carvalho Aguiar 44,
5º andar, Bloco II
(secretaria de pneumologia),
05403-900 São Paulo, Brazil.
E-mail: leticiakawano@gmail.com

Received on August 5, 2014; accepted in revised form on December 19, 2014.

Competing interests: none declared.

Introduction

Mixed connective tissue disease (MCTD) is a systemic autoimmune disorder that is characterised by a combination of findings usually observed in systemic lupus erythematosus, scleroderma, and polymyositis. A high titer of anti-nuclear ribonucleoprotein (RNP) in the presence of these symptoms appears to be unique to MCTD (1-3).

A growing body of evidence indicates that interstitial lung disease (ILD) is highly prevalent among MCTD patients. ILD and pulmonary arterial hypertension (PAH) are serious complications of MCTD associated with increased mortality, and a subset of these patients will present with severe pulmonary fibrosis and increased mortality (4-6). However, little is known about the pattern of long-term progression of ILD in MCTD. There are only a few studies that provide longitudinal information regarding ILD in MCTD (5-8), and to the best of our knowledge, none of them have included a quantitative analysis of changes in pulmonary functional parameters or imaging results. Consequently, the parameters that are most sensitive for detecting the progression of ILD in MCTD and for determining the extent of progression have not been identified.

Therefore, the present study had three aims: i) to describe the most common abnormalities observed in pulmonary function tests (PFTs) and high resolution computed tomography (HRCT) scans of the chest in long-term MCTD patients; ii) to measure the variation over 10 years in PFT results and HRCT findings (semi-quantitative and qualitative analysis); and iii) to identify functional and imaging correlations.

Materials and methods

Patients and study design

In this retrospective cohort study, we systematically evaluated 53 unselected consecutive MCTD patients who were first evaluated 10 years before (9) at the tertiary rheumatology outpatient clinic of the Hospital das Clinicas, University of Sao Paulo, Brazil and who had been stringently diagnosed according to the Kasukawa's criteria (1). Follow-up PFTs, chest HRCT images and structured clinical interviews were performed. Informed consent was obtained from all participants. The study protocol was approved by the local research ethics committee (n. 0099/11). The data obtained 10 years before were considered the baseline against which the follow-up data were compared. Clinical data retrospectively extracted from medical records were:

1) smoking status, with active or ex-smokers defined as having a minimum of one cigarette a day for a minimum of one year;

2) presence of secondary Sjögren syndrome;

3) PAH, which was defined by right heart catheterisation performed in patients with pulmonary artery systolic pressure >40 mmHg at an annual screening echocardiogram, in accordance with the standard of care in our institution;

4) autoantibody profile including anti-nuclear (ANA), anti-ribonucleoprotein (RNP), anti-Sm, anti-SSA/Ro, anti-SSB/La, anti-topoisomerase I (Scl-70), anti-centromere (ACA), anti-double-stranded DNA (dsDNA), anti-histidyl-tRNA synthetase (anti-Jo1), and IgG and IgM anticardiolipin (aCL) autoantibodies and rheumatoid factor (RF);

5) drug treatment, defined as treatment for any manifestation of MCTD at any time in the 10-year period and consisting of prednisone or equivalent >5 mg/day (usually in the range of 5 mg to 1 mg/kg/day) or of cyclophosphamide, azathioprine, or methotrexate with or without corticosteroids.

For those patients who died or were lost to follow-up, the last clinical, functional, and radiologic data available were retrieved from the medical records.

The patients received standard medical care at the discretion of the attending physician during the 10-year period.

Pulmonary function tests

Spirometry (Medical Graphics Corporation [MGC], St Paul, MN, USA), plethysmographic lung volume measurements, and diffusion capacity for carbon monoxide (DL_{CO}) (Elite Dx, Elite Series™, MGC) were used to assess lung function. All measurements were obtained according to the recommended standards (10-12). Reference

values were derived from the Brazilian population (13-15).

High-resolution computed tomography protocols

HRCT scans were obtained within a maximum interval of six months from the PFTs. Scans at baseline had been obtained in a single-slice scanner (HiSpeed Advantage, GE Medical Systems, Milwaukee, WI, USA), and the follow-up images were acquired in multi-slice systems (Philips Brilliance 10 or 16, Philips Medical Systems, Cleveland, OH, USA). Scans were performed from the lung apices to the bases without intravenous contrast medium. Images were obtained with patients in the supine position during breath-holding in deep inspiration; whenever necessary, prone scans were obtained. The images were acquired with a field of view of 36 cm, 512×512 matrix, 120 kVp, and 150–200 mA using helical mode in the multi-slice systems and a step-and-shoot mode in the single-slice scanner. In both cases, the images were reconstructed using a high spatial resolution kernel with 1.0 mm thickness and 10 mm reconstruction intervals. All images were displayed in two fixed windows; lung windows were set to –700 Hounsfield Units (HU) with 1,500 HU width, while mediastinal windows had a centre of 20 HU and width of 400 HU.

Two chest radiologists who were blinded to the clinical and functional data independently reviewed the chest scans. The images were presented in random order and scored for each pattern of lung abnormality as defined elsewhere (16), and the presence of oesophageal dilatation was also noted (17).

Findings of ground-glass opacities, reticulation, honeycombing, and bronchiolectasis were considered evidence of ILD. The radiologists scored each lobe for the extent of ILD (from 0 to 100%) to the nearest 5%, counting the lingula as a separate lobe, yielding a total ILD score (total %ILD-HRCT score) (18). The scores obtained by the two radiologists were averaged, except in cases of discrepancies of >20% between readings, which were resolved by consensus. The average %ILD-HRCT score for the right and left lower lobes, referred to as the lower-lobes %ILD-HRCT score, was also recorded and analysed, because ILD in MCTD predominantly affects the lower lobes (19,20).

Data analysis

Data were reported as mean ± standard deviation (SD) for variables with normal distribution or as the median ($IQ_{25–75\%}$) for variables with non-normal distribution. Unpaired *t*-tests or the Mann-Whitney U-test were used to compare continuous variables between different subgroups. Paired *t*-tests or Wilcoxon tests were used to compare within-subject results. Categorical variables were compared by Fisher's exact or Chi-square test. Spearman's correlation coefficient was used to evaluate the correlation between functional variables and imaging scores. Differences were considered significant at $p<0.05$. The data were analysed with SigmaStat version 3.5 (Systat Software, Inc., San Jose, CA).

Results

From the original unselected sample of 53 MCTD patients, 7 (13%) were lost to follow up, 4 (7.5%) had their diagnosis changed, and 3 (5.6%) died. Therefore, the study included 39 patients. MCTD manifestations according to the Kasukawa criteria and the general characteristics of the patients are presented in Tables I and II, respectively.

The 7 (13%) patients lost to follow-up were followed-up for a median time of 7 years (range: 0.7–9 years) and their baseline functional parameters did not differ from those of the rest of the sample: the mean baseline forced vital capacity (FVC) was 72% of predicted and the mean baseline DL_{CO} was 86% of predicted (p = ns); the final FVC retrieved from patient records was stable in 6 of these patients; in one patient there was no information regarding follow-up PFTs.

The causes of death in the 3 patients who died during the study period were sepsis secondary to a skin infection, decompensated left heart failure, and thrombocytopenic thrombotic purpura. At the baseline evaluations, the median DL_{CO} of these 3 patients was 60% of predicted and the median FVC was 69% of predicted. At the last recorded evaluation before death, the mean disease duration was 17 years and the mean age was 45.3 years. One of these patients had a normal HRCT at the final evaluation and 2 showed signs of ILD (ground-glass opacities, bronchiolectasis and reticulation) with mild (10%) and extensive (30%) involvement, respectively (10% and 30% lower-lobes %ILD-HRCT scores).

Two of the 4 patients who had changed diagnoses were moved to overlap syndromes of systemic lupus erythematosus and scleroderma (n=1) and systemic lupus erythematosus and rheumatoid arthritis (n=1); the other 2 fulfilled the criteria for isolated scleroderma and isolated systemic lupus erythematosus.

Table I. MCTD manifestations acccording to the Kasukawa criteria*.

Characteristic	n=39
A. *Common symptoms*	
Raynaud's phenomenon	38 (97%)
Swollen fingers or hands	33 (85%)
B. *Anti-RNP*	37 (95%)¥
C. *Mixed symptoms*	
SLE-like findings	
• polyarthritis	28 (71%)
• lymphadenopathy	7 (18%)
• facial erythema	12 (31%)
• pericarditis or pleuritis	4 (10%)
SScl-like findings	
• sclerodactyly	14 (36%)
• pulmonary fibrosis, restrictive changes in PFTs or reduced DL_{CO} ‖	30 (77%)
• oesophageal hypomotility or dilatation	35 (90%)
PM-like findings	
• muscle weakness	14 (36%)
• elevated serum levels of muscle enzymes	16 (41%)
• myogenic pattern on EMG	2 (5%)

Data are presented as absolute number of patients and percentage.

*Kasukawa criteria for MCTD requires at least one of the two common symptoms plus positive for anti-RNP plus one or more of the mixed symptoms in at least two of the three (SLE-like, SScl-like or PM-like) disease categories. (1)

¥At diagnosis, all patients were anti-RNP positive at high titers (>1:1000) detected by haemagglutination. Two patients became anti-RNP negative over the time.

‖At this study, pulmonary fibrosis was identified with chest HRCT.

anti-RNP: ribonucleoprotein antibody; SLE: systemic lupus erythematosus; SScl: systemic sclerosis; PM: polymyositis; PFTs: pulmonary function tests; DL_{CO}: diffusion capacity for carbon monoxide; EMG: electromyogram.

Table II. Follow-up characteristics of MCTD patients.

Characteristic	n=39
Female sex	39 (100%)
Mean age, years	53 ± 11.3
Median duration of MCTD symptoms, years *	19 (15-28)
Active smokers	1 (2.5%)
Ex-smokers	11 (28%)
Sjögren syndrome	5 (13%)
Pulmonary arterial hypertension¥	2 (5%)
Treated patients ‖	37 (95%)
Dyspnea	19 (49%)
Chronic cough	16 (41%)
Autoantibodies:	
ANA titer > 1/160	39 (100%)
Anti-DNA_{ds}	0 (0%)
Anti-RNP ¶	37 (95%)
Anti-Sm	1 (2%)
Anti-Ro/SSA	10 (26%)
Anti-La/SSB	1 (2.5%)
aCL IgG	0 (0%)
aCL IgM	1 (2%)
RF	10 (26%)

Data are presented as absolute values and percentages, mean ± SD and median and $IQ_{25\text{-}75\%}$.
*Median duration of MCTD symptoms, except Raynaud phenomenon.
¥Pulmonary arterial hypertension as defined in by right heart catheterisation performed in patients with pulmonary artery systolic pressure >40 mmHg in annual screening echocardiogram
‖A treated patient is defined as any patient who at any time during the 10-year period received 5 mg/day or more of prednisone or its equivalent in isolation or in combination with cyclophosphamide, azathioprine, and/or methotrexate. Patients who received only chloroquine diphosphate or hydroxychloroquine sulfate were not included in the treated group.
¶All patients were anti-RNP positive at high titers (>1:1000) detected by haemagglutination at diagnosis, but two patients became anti-RNP negative over the 10-year period. None of the patients were positive for anti-topoisomerase (anti-Scl70), anti-centromere (ACA) or anti-histidyl-tRNA synthetase (anti-Jo1) antibodies.
ANA: antinuclear antibody; RNP: ribonucleoprotein antibody; aCL: anticardiolipin antibodies; RF: rheumatoid factor.

Table III. Changes in pulmonary function tests in MCTD patients after ten years.

Variables	Baseline	Follow-up	*p*-value
FVC (L)*	2.58 ± 0.54	2.45 ± 0.56	0.09
%predicted	77% ± 15	78% ± 16	0.78
FEV_1 (L)*	2.13 ± 0.44	1.96 ± 0.42	*p*<0.001
%predicted	83% ± 16	81% ± 20	0.52
TLC (L)¥	4.00 ± 0.63	3.92 ± 0.72	0.44
%predicted	86% ± 15	83% ± 14	0.34
DL_{CO} (ml/min/mmHg)¥	17.7 ± 5.1	15.1 ± 4.3	*p*=0.006
%predicted	84% ± 21	71% ± 19	*p*<0.001

Data are presented in absolute values and as percent of the predicted value, mean ± SD. Predicted values have changed in follow-up measures due to ageing of the patients.
*n=36 for FVC and FEV_1. ¥n=33 for TLC and DL_{CO}.
L: liters; FVC: forced vital capacity; FEV_1: forced expiratory volume in the first second; TLC: total lung capacity; DL_{CO}: single breath carbon dioxide diffusing capacity.

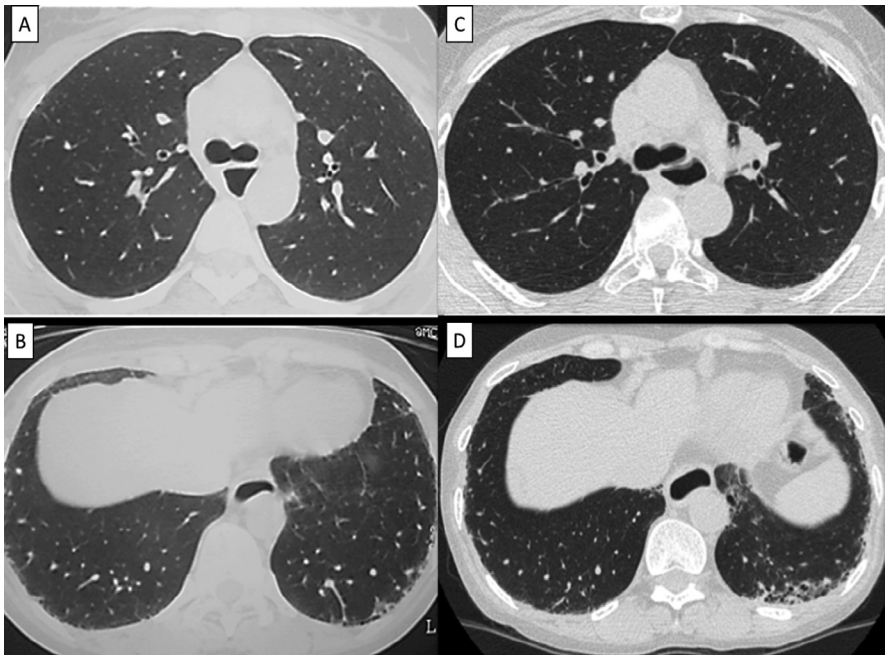

Fig. 1. The most prevalent image findings in MCTD were ground-glass opacities and reticulation, predominately in the periphery of the lower lobes. Slides **C** and **D** show the same patient as slides **A** and **B**, after 10 years. There is mild progression of disease with an increase in signs of fibrosis, (traction bronchiolectasis and an elevated left diaphragmatic dome). The baseline lower-lobes %ILD-HRCT score was 7.5% and the follow-up score was 12.5%.

One of the 39 included patients had existing PAH at baseline, and during the 10-year follow-up period, 6 (15%) patients were evaluated by right heart catheterisation after the annual screening echocardiogram indicated an increased estimated pulmonary systolic arterial pressure, but only 1 additional (2.5%) patient had the diagnosis of PAH confirmed.

Nineteen (49%) patients presented with dyspnea, 2 with more severe dyspnea (modified Medical Research Council Dyspnea Scale [MMRC] (3-4).

All patients were in stable condition when PFTs and HRCT were performed.

Pulmonary function tests

Of the 39 included patients, 3 were unable to perform FVC and forced expiratory volume in 1 second (FEV_1) measurements and 6 were unable to perform DL_{CO} and total lung capacity (TLC) tests. Therefore, FVC and FEV_1 were measured in 36 (92%) patients and DL_{CO} and TLC were measured in 33 (84%) patients. The characteristics of the patients who were unable to perform follow-up PFTs did not differ from the rest of the patient population (mean age 54.3 years, mean disease duration 20.4 years, mean baseline FVC 82% of predicted).

Of the 33 patients, 17 (51.5%) had abnormal PFTs at baseline, but only 2 had FVC values of <50% of predicted.

A statistically significant relative decrease of 15% in mean DL_{CO} was detected after 10 years (from 84% to 71% of predicted, *p*<0.001) (Table III), and there were 4 (12%) patients who had relative decreases in DL_{CO} of >40%.

The FVC was slightly reduced at baseline (77% of predicted) but remained stable throughout the 10-year period; 3 (8%) patients had relative decreases in FVC of >20%. Baseline FEV_1 and TLC values were within the normal range and remained stable at follow-up.
Although 30% of the patients were ex-smokers, an obstructive ventilatory defect (FEV_1/FVC = 0.67) was observed in only one, non-smoking, patient.

High-resolution computed tomography
Two of the 39 included patients did not undergo HRCT of the chest; 37 (94%) patients had a follow-up chest HRCT, and 35 (89%) patients had both baseline and follow-up images available for analysis.
Eight (23%) patients had normal baseline HRCT scans (total %ILD-HRCT score = 0). In the remaining patients, peripheral ground-glass opacities and reticulation, predominating in the lower lobes, were the most common findings (Fig. 1). The lower-lobes %ILD-HRCT scores, but not the total %ILD-HRCT scores, were able to identify statistically significant changes between baseline and follow-up images. The median baseline lower-lobes %ILD-HRCT score was 7.5% ($IQ_{25–75\%}$ 0.6–17.5%) and progressed to 11.2% ($IQ_{25–75\%}$ 3.7–33.8%) at 10 years (p=0.02) (Table IV). Signs of fibrosis, *i.e.* honeycombing and traction bronchiolectasis, increased, as did the prevalence of oesophageal dilatation (68% to 90%, p=0.029) (Table IV). Of note, 7 (20%) patients had total %ILD-HRCT scores of >20% at baseline (mean total %ILD-HRCT score 29%), but, interestingly, none of these 7 patients exhibited increased total or lower-lobes %ILD-HRCT scores at follow-up.

Correlation between pulmonary function tests and high resolution computed tomography
A statistically significant moderate negative correlation was observed between baseline and follow-up FVC vales and lower-lobes %ILD-HRCT scores and between the baseline and follow-up DL_{CO} values and lower-lobes %ILD-HRCT scores (Table V). Because DL_{CO} was the only functional parameter that changed significantly during the follow-up period, we chose to evaluate the correlation of the 10-year change (Δ) in DL_{CO} with the 10-year change (Δ) in the lower-lobes %ILD-HRCT score, and these parameters also showed a moderate negative correlation (Table V; Fig. 2).

Table IV. Prevalence and evolution over ten years of imaging findings in MCTD patients.

Characteristics	Baseline*	Follow-up*	*p*-value
Lower-lobes %ILD-HRCT score¥	7.5% (0.6 – 17.5)	11.25% (3.7 – 33.8)	0.02
Image findings ‖			
Ground-glass opacities	74%	77%	1.0
Reticulation	65%	77%	0.43
Traction bronchiolectasis	32%	58%	0.041
Honeycombing	13%	45%	0.005
Nodules	6%	6%	1.0
Consolidation	0%	0%	1.0
Pleural thickening	3%	3%	1.0
Pleural effusion	0%	3%	1.0
Emphysema	0%	11%	0.11
Oesophageal dilatation	68%	90%	0.029

*Number of patients = 35. ¥Extent of disease quantified by lower-lobes %ILD-HRCT score; median values with $IQ_{25–75\%}$ in parentheses. ‖Data are presented as the percentage of patients with the specified finding.

Table V. Correlation between functional parameters and quantified imaging results (lower-lobes %ILD-HRCT score.

Variables*	Coefficient¥	*p*-value
Baseline		
FVC and lower-lobes %ILD-HRCT	-0.49	0.005
DL_{CO} and lower-lobes %ILD-HRCT	-0.53	0.003
Follow-up		
FVC and lower-lobes %ILD-HRCT	-0.51	0.003
DL_{CO} and lower-lobes %ILD-HRCT	-0.62	<0.001
Δ DL_{CO} and Δ lower-lobes %ILD-HRCT ‖	-0.39	0.034

*For correlation with FVC, n=33. For correlations with DL_{CO}, n=29. ¥Spearman correlation coefficient. ‖ Δ DL_{CO}: follow-up DL_{CO} minus baseline DL_{CO}; Δ lower-lobes %ILD-HRCT: follow-up score minus baseline score.
FVC: forced vital capacity; DL_{CO}: single breath carbon dioxide diffusing capacity; lower-lobes %ILD-HRCT: score quantifying tomographic image findings in the lower lobes.

Discussion

In this 10-year retrospective analysis of a treated unselected sample of long-term MCTD patients, we found a high prevalence of functional and imaging changes that were suggestive of ILD. We found that FVC, FEV_1, and TLC values remained stable, while a 15% relative decrease in DL_{CO} was detected. The most prevalent imaging findings were peripheral ground-glass opacities and reticulation, which were predominately in the lower lobes and of mild extent, suggestive of a non-specific interstitial pneumonia (NSIP) pattern. After 10 years, the prevalence of HRCT findings suggestive of fibrosis, *i.e.* honeycombing and traction bronchiolectasis, and of oesophageal dilatation, increased. Interestingly, at the first study with this sample of patients lower DL_{CO} and an increased prevalence of ILD on HRCT were more common in patients with oesophageal dilatation (9). However, at follow-up evaluation, the prevalence of oesophageal dilatation did not differ between the functional deterioration subgroup and functionally stable/improved subgroup (data not shown). ILD, gastroesophageal reflux (GER), and oesophageal motor impairment are very common and frequently coexist in patients with MCTD. The nature of the association between oesophageal involvement and ILD in MCTD is not completely understood, and further studies are necessary to

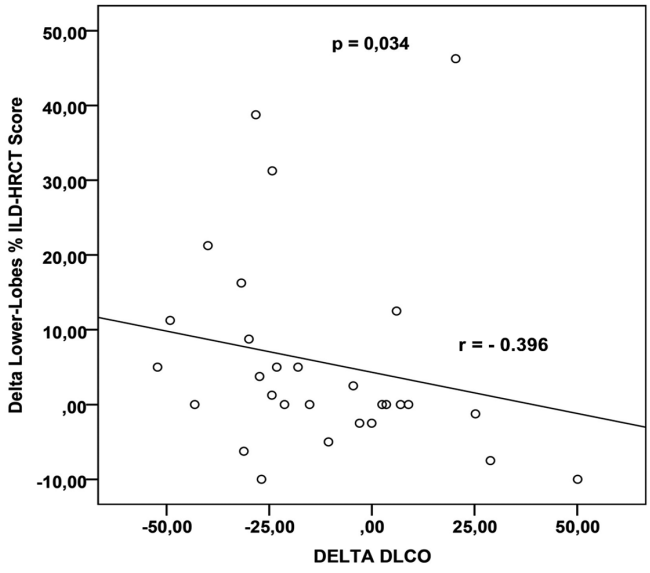

Fig. 2. Moderate correlation between the 10-year change (Δ) in DL_{CO} and the 10-year change (Δ) in lower-lobes %ILD-HRCT scores.

clarify such issue. Meanwhile, one may speculate either a causal relationship: GER promoting and aggravating ILD or either they are both manifestations of the scleroderma-like findings of MCTD and therefore come together.

Few longitudinal studies analysing ILD in MCTD have been undertaken (5-8), and to our knowledge, ours is the first study in which the long-term progression of HRCT findings and PFT trends were both quantified. The general stability of the PFT results among the patients in our sample, among whom only a slight decrease in DL_{CO} was observed, together with a 5.6% mortality rate, agrees with the evidence in the literature that in general, the ILD of MCTD has a more benign course than other ILDs such as scleroderma or idiopathic pulmonary fibrosis (21-25). However, the stability observed in our study might be in part related to the characteristics of the study population, which was mainly composed of patients with long-term disease. At the follow-up analysis, the mean disease duration was 19 years, indicating that this population had a mean disease duration of 9 years at baseline. In scleroderma patients with ILD, the most rapid decline in FVC occurs within three years of disease onset, indicating that lung injury and fibrosis are early complications (26).

In our cohort, the mild morphologic changes and the mild disease progression observed on HRCT may explain why DL_{CO} was a marker of disease progression while FVC was not: DL_{CO} appears to be a more sensitive functional parameter in general in terms of detection and follow-up of diffuse ILD (27-33), and in one recent study of MCTD patients, FVC remained at 80% of the predicted value even among those with HRCT findings of more severe disease, while the DL_{CO} was decreased (4). The low prevalence of PAH in our population gives further support to the use of the observed decrease in DL_{CO} as a surrogate marker of disease progression, and it is also in agreement with recent findings of phenotypic clusters in MCTD in which patients with ILD and PAH did not cluster together (34). As such, a patient population with a high prevalence of ILD, such as that observed in our study, would not be expected to have a high prevalence of PAH.

Although both DL_{CO} and qualitative/quantitative image findings detected the mild progression of ILD, there was only a moderate correlation between the functional variables and the extent of disease on HRCT, either as absolute values or as Δ DL_{CO} and Δ lower-lobes %ILD-HRCT. Indeed, a moderate correlation between PFTs and extent of disease on HRCT is not uncommonly reported for ILDs in general (35), and only few studies have reported more robust correlations (18, 27). It is unlikely that the moderate (and not good) correlation between pulmonary function parameters and image scores was related to the maximum accepted interval of 6 months between follow-up PFTs and follow-up HRCT, because most of the HRCT exams were actually performed within a month of the PFTs, and the ILD in our study population was not rapidly progressive. Another possible explanation for the moderate correlation between DL_{CO} and the radiographic extent of disease is that the DL_{CO} may be a more sensitive indicator of microscopic ILD before it can be detected macroscopically by changes in HRCT images. Therefore, morphologic and functional evaluations, when conducted together, seem to be additive in improving accuracy. Lastly, although not directly relevant to our study, a high prevalence of PAH could decrease the correlation between DL_{CO} and HRCT, because PAH would not be detected on interstitial image analysis, despite its association with significant decreases in DL_{CO}.

A theoretical drawback in using DL_{CO} as a tool to evaluate progression of ILD is the variability in the measurements. However, clinically acceptable reproducibility has previously been demonstrated with the use of a standardised protocol (10, 15). In addition, the natural decrease in the predicted values of DL_{CO} over a 10-year span in the general Brazilian population is approximately 4%, which is less than the 15% decrease observed in our study. Furthermore, the decrease in DL_{CO} retained statistical significance when the % predicted DL_{CO} was analysed and the effect of ageing was already taken into account (15). Therefore, the analysis of DL_{CO} trends can be a useful method for evaluating ILD progression in MCTD, even during long-term follow-up, and in fact, DL_{CO} was the most sensitive functional parameter in this study.

Despite the strength of our findings, our study has limitations. The loss to follow-up of 13% of patients may theoretically have introduced a selection bias. However, even these patients were followed for a median time of 7 years, and their baseline and final follow-up FVC values did not differ from the rest of the cohort. Thus, we have reason to believe that their disease had a behavior similar to that of the rest of the cohort. Nevertheless, we cannot confirm that they are still alive since we lost contact with

them. Another limitation is that the impact of treatment on our observations was not subject to analysis. However, our observations that trends in DL_{CO} and qualitative and quantitative imaging analysis are the most sensitive parameters to detect subtle progression of ILD in MCTD are not invalidated by the fact that treatment effects were not evaluated. Additionally, because we have only two sets of PFTs and HRCT images, collected 10 years apart, we cannot know whether the patients had a slowly progressive deterioration over the 10 years or whether short periods of rapid disease progression occurred in an otherwise stable disease state.

This is the first time that PFT and HRCT image findings have been objectively quantified simultaneously in a population of MCTD patients. In summary, after comparing baseline and follow-up PFT and HRCT results over a 10-year interval in an unselected sample of long-term MCTD patients, we observed that functional and radiologic alterations suggestive of ILD are prevalent, relatively mild, and slightly progressive over time. Because we observed larger decreases in PFT parameters and more extensive disease on HRCT in a minority of patients, and because an increased extent of ILD in MCTD patients has already been associated with increased mortality (4), the importance of long-term follow-up for MCTD patients with pulmonary involvement cannot be overstated. The present study indicates that the most sensitive parameters for detecting subtle progression of ILD in MCTD patients are trends in DL_{CO}, quantification of the extent of disease in the lower-lobes by HRCT, and qualitative analysis of HRCT findings.

Acknowledgements

We are particularly grateful for the valuable help with data acquisition provided by Renata Miossi.

References

1. KASUKAWA R: Mixed connective tissue disease. *Intern Med*. 1999; 38: 386-93.
2. SHARP GC, IRVIN WS, TAN EM, GOULD RG, HOLMAN HR: Mixed connective tissue disease--an apparently distinct rheumatic disease syndrome associated with a specific antibody to an extractable nuclear antigen (ENA). *Am J Med* 1972; 52: 148-59.
3. SHARP GC, HOFFMAN RW: Clinical, immunologic, and immunogenetic evidence that mixed connective tissue disease is a distinct entity: comment on the article by Smolen and Steiner. *Arthritis Rheum* 1999; 42: 190-1; reply 3-6.
4. GUNNARSSON R, AALØKKEN TM, MOLBERG Ø *et al*.: Prevalence and severity of interstitial lung disease in mixed connective tissue disease: a nationwide, cross-sectional study. *Ann Rheum Dis* 2012; 71: 1966-72.
5. BURDT MA, HOFFMAN RW, DEUTSCHER SL, WANG GS, JOHNSON JC, SHARP GC: Long-term outcome in mixed connective tissue disease: longitudinal clinical and serologic findings. *Arthritis Rheum* 1999; 42: 899-909.
6. BODOLAY E, SZEKANECZ Z, DÉVÉNYI K *et al*.: Evaluation of interstitial lung disease in mixed connective tissue disease (MCTD). *Rheumatology* (Oxford) 2005; 44: 656-61.
7. IZUMIYAMA T, HIDA W, ICHINOSE M *et al*.: Small airway involvement in mixed connective tissue disease. *Tohoku J Exp Med* 1993; 170: 273-83.
8. SULLIVAN WD, HURST DJ, HARMON CE *et al*.: A prospective evaluation emphasizing pulmonary involvement in patients with mixed connective tissue disease. *Medicine* (Baltimore) 1984; 63: 92-107.
9. FAGUNDES MN, CALEIRO MT, NAVARRO-RODRIGUEZ T *et al*.: Esophageal involvement and interstitial lung disease in mixed connective tissue disease. *Respir Med* 2009; 103: 854-60.
10. MACINTYRE N, CRAPO RO, VIEGI G *et al*.: Standardisation of the single-breath determination of carbon monoxide uptake in the lung. *Eur Respir J* 2005; 26: 720-35.
11. MILLER MR, HANKINSON J, BRUSASCO V *et al*.: Standardisation of spirometry. *Eur Respir J* 2005; 26: 319-38.
12. WANGER J, CLAUSEN JL, COATES A *et al*.: Standardisation of the measurement of lung volumes. *Eur Respir J* 2005; 26: 511-22.
13. PEREIRA CA, SATO T, RODRIGUES SC: New reference values for forced spirometry in white adults in Brazil. *J Bras Pneumol* 2007; 33: 397-406.
14. NEDER JA, ANDREONI S, CASTELO-FILHO A, Nery LE. Reference values for lung function tests. I. Static volumes. *Braz J Med Biol Res* 1999; 32: 703-17.
15. NEDER JA, ANDREONI S, PERES C, NERY LE: Reference values for lung function tests. III. Carbon monoxide diffusing capacity (transfer factor). *Braz J Med Biol Res* 1999; 32: 729-37.
16. HANSELL DM, BANKIER AA, MACMAHON H, MCLOUD TC, MÜLLER NL, REMY J: Fleischner Society: glossary of terms for thoracic imaging. *Radiology* 2008; 246: 697-722.
17. BHALLA M, SILVER RM, SHEPARD JA, MCLOUD TC: Chest CT in patients with scleroderma: prevalence of asymptomatic esophageal dilatation and mediastinal lymphadenopathy. *Am J Roentgenol* 1993; 161: 269-72.
18. MARTEN K, DICKEN V, KNEITZ C *et al*.: Interstitial lung disease associated with collagen vascular disorders: disease quantification using a computer-aided diagnosis tool. *Eur Radiol* 2009; 19: 324-32.
19. KHANNA D, TSENG CH, FARMANI N *et al*.: Clinical course of lung physiology in patients with scleroderma and interstitial lung disease: analysis of the Scleroderma Lung Study Placebo Group. *Arthritis Rheum* 2011; 63: 3078-85.
20. KOZUKA T, JOHKOH T, HONDA O *et al*.: Pulmonary involvement in mixed connective tissue disease: high-resolution CT findings in 41 patients. *J Thorac Imaging* 2001; 16: 94-8.
21. SAITO Y, TERADA M, TAKADA T *et al*.: Pulmonary involvement in mixed connective tissue disease: comparison with other collagen vascular diseases using high resolution CT. *J Comput Assist Tomogr* 2002; 26: 349-57.
22. FERRI C, VALENTINI G, COZZI F *et al*.: Systemic sclerosis: demographic, clinical, and serologic features and survival in 1,012 Italian patients. *Medicine* (Baltimore) 2002; 81: 139-53.
23. BJORAKER JA, RYU JH, EDWIN MK *et al*.: Prognostic significance of histopathologic subsets in idiopathic pulmonary fibrosis. *Am J Respir Crit Care Med* 1998; 157: 199-203.
24. KING TE, BEHR J, BROWN KK *et al*.: BUILD-1: a randomized placebo-controlled trial of bosentan in idiopathic pulmonary fibrosis. *Am J Respir Crit Care Med* 2008; 177: 75-81.
25. RAGHU G, BROWN KK, BRADFORD WZ *et al*.: A placebo-controlled trial of interferon gamma-1b in patients with idiopathic pulmonary fibrosis. *N Engl J Med* 2004; 350: 125-33.
26. PLASTIRAS SC, KARADIMITRAKIS SP, ZIAKAS PD, VLACHOYIANNOPOULOS PG, MOUTSOPOULOS HM, TZELEPIS GE: Scleroderma lung: initial forced vital capacity as predictor of pulmonary function decline. *Arthritis Rheum* 2006; 55: 598-602.
27. WELLS AU, HANSELL DM, RUBENS MB *et al*.: Fibrosing alveolitis in systemic sclerosis: indices of lung function in relation to extent of disease on computed tomography. *Arthritis Rheum* 1997; 40: 1229-36.
28. BOUROS D, WELLS AU, NICHOLSON AG *et al*.: Histopathologic subsets of fibrosing alveolitis in patients with systemic sclerosis and their relationship to outcome. *Am J Respir Crit Care Med* 2002; 165: 1581-6.
29. LATSI PI, DU BOIS RM, NICHOLSON AG *et al*.: Fibrotic idiopathic interstitial pneumonia: the prognostic value of longitudinal functional trends. *Am J Respir Crit Care Med* 2003; 168: 531-7.
30. SCHMIDT SL, TAYOB N, HAN MK *et al*.: Predicting pulmonary fibrosis disease course from past trends in pulmonary function. *Chest* 2014; 145: 579-85.
31. MARTINEZ FJ, FLAHERTY K: Pulmonary function testing in idiopathic interstitial pneumonias. *Proc Am Thorac Soc* 2006; 3: 315-21.
32. WELLS AU, KING AD, RUBENS MB, CRAMER D, DU BOIS RM, HANSELL DM: Lone cryptogenic fibrosing alveolitis: a functional-morphologic correlation based on extent of disease on thin-section computed tomography. *Am J Respir Crit Care Med* 1997; 155: 1367-75.
33. BALDI BG, PEREIRA CA, RUBIN AS *et al*.: Highlights of the Brazilian Thoracic Association guidelines for interstitial lung diseases. *J Bras Pneumol* 2012; 38: 282-91.
34. SZODORAY P, HAJAS A, KARDOS L *et al*.: Distinct phenotypes in mixed connective tissue disease: subgroups and survival. *Lupus* 2012; 21: 1412-22.
35. GOH NS, DESAI SR, VEERARAGHAVAN S *et al*.: Interstitial lung disease in systemic sclerosis: a simple staging system. *Am J Respir Crit Care Med* 2008; 177: 1248-54.