Evolution of imaging findings, laboratory and functional parameters in rheumatoid arthritis patients after one year of treatment with anti-TNF-α agents

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Abstract

Objective

To investigate the efficacy and safety of anti-TNF-α agent treatment compared to non-biologic DMARDs in rheumatoid arthritis patients.

Methods

82 consecutive patients, 29 males, 53 females, aged 42–79, diagnosed with RA and suitable for anti-TNF-α treatment composed two study groups: 42 with pre-existing rheumatoid arthritis-related interstitial lung disease (RA-ILD) and 40 without RA-ILD. Respective control groups consisted of 44 patients with pre-existing RA-ILD and 44 patients without RA-ILD, treated with non-biologic DMARDs. All patients underwent chest high resolution computed tomography (HRCT), pulmonary function tests (PFTs) and peripheral blood biomarkers at baseline and after one year of treatment.

Results

There was a significant decrease of air trapping extent and bronchial wall thickening after treatment in RA-ILD and RA-non ILD study groups ($p<0.05$). This was accompanied by a statistically significant improvement of maximum mid-expiratory flow ($MMEF_{75-25}$), RV and RV/TLC in both study groups ($p<0.05$). In the RA-ILD study group ILD extent scores remained unchanged after anti-TNF-α treatment. None of the RA-non ILD group developed new-onset ILD. In both RA-ILD and RA-non ILD control groups, HRCT findings and PFTs did not differ significantly at the one-year follow-up study. Methotrexate (MTX) regression analysis showed in both RA-ILD study and control groups a negative correlation between MTX dose and ILD extent score at one-year and between MTX dose and air trapping extent at baseline and after one year of treatment.

Conclusion

Anti-TNF-α treatment, in contrast to non-biologic DMARDs, there was an improvement of small airways disease. There was no new-onset ILD or exacerbation of preexisting-ILD, especially in patients treated with anti-TNF-α agents, supporting the efficacy and favourable safety profile of this treatment in RA patients.

Key words

RA-ILD, anti-TNF-α, HRCT, PFTs, non-biologic DMARDs

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Introduction

Lung involvement in rheumatoid arthritis (RA) has been identified in early symptomatic disease and occasionally prior to the onset of articular symptoms (1-7). Rheumatoid arthritis-associated interstitial lung disease (RA-ILD) is the most common pulmonary manifestation observed in 50% of chest CT and in 5% of chest radiography and is associated with ≥20% morbidity in patients with prevalent disease (8-13).

Airway involvement in RA patients has been less emphasised in the literature comparing to ILD, with a prevalence reaching 51% in symptomatic patients (14-16). Small airways disease has been reported in up to two-thirds of RA patients with a high prevalence, even among those without ILD (17). High resolution computed tomography (HRCT) scans appear to be more sensitive compared to pulmonary function tests (PFTs) for small airways disease detection (17, 18).

There is controversy regarding the effects of TNF-α targeted therapy on the lung. Some studies report stability and others new-onset or progression of RA-ILD following anti-TNF-α treatment (19-25). This prospective study aims to evaluate the effect of TNF-α inhibitors on airways and lung parenchyma compared to non-biological disease-modifying anti-rheumatic drugs (nbDMARDs), with regard to efficacy and safety.

Materials and methods

This prospective study was conducted from January 2011 to August 2015 according to the principles of the Helsinki Declaration and approved by the Institutional Ethics Committee. All patients gave written informed consent. Inclusion criteria for patients in the study group consisted of diagnosed rheumatoid arthritis (RA) according to the revised classification criteria for RA of the American College of Rheumatology and the European League Against Rheumatism, refractory to conventional treatment with nbDMARDs and therefore candidates for anti-TNF-α agent therapy (25, 26). Exclusion criteria included history of asthma, primary pulmonary hypertension, left heart failure and exposure to silica.

Eighty-two (82) patients, 29 males, 53 females, of mean age 60 (range: 42–79 years) were prospectively recruited. Pre-existing RA-ILD (RA-ILD group) was present in 42/82, diagnosed on the basis of clinical findings, pulmonary function tests (PFTs) and chest HRCT findings. The remaining 40/82 had no imaging or clinical findings of ILD (RA-non ILD group). Twenty (20/82) patients were active smokers, 12 patients in the RA-ILD positive and 8 in the RA-ILD negative group, with a mean cigarette consumption of 22 pack/years.

A control group of eighty-eight (88) RA patients diagnosed with the same inclusion criteria and following the same exclusion criteria who underwent (nb) DMARD treatment were divided into two control groups: 44 patients with pre-existing RA-ILD and 44 without RA-ILD. Patients' demographics are shown in Table I.

All patients underwent paired inspiratory and expiratory chest HRCT, PFTs and laboratory tests at baseline and following 1-year treatment. Assessment of clinical response of RA was performed using Disease Activity Score 28 (DAS 28) (27). PFTs, laboratory tests and DAS 28 were performed on average within 2 days (mean, 1.71±5.9) of obtaining chest HRCT scans.

Treatment

Eighty-two RA patients received anti-TNF-α treatment: 68 received infliximab (recommended dose of 3 mg/kg as an intravenous regimen at 0, 2 and 6 weeks, followed by a maintenance regimen of 3 mg/kg every 8 weeks thereafter), 10 patients received etanercept (50 mg, weekly) and 4 patients adalimumab (40 mg, weekly). Anti-TNF-α treatment in all above patients was combined with low-dose methotrexate (MTX) (7.5 mg per os/week).

Eighty-eight control group patients received nbDMARD treatment, among them 68 MTX alone (mean dose of 15 mg per os/week), 20 hydroxychloroquinine (mean dose 400 mg per os/day) combined with low-dose MTX (7.5 mg per os/week).

High-resolution CT

All patients underwent chest HRCT

exam consisting of paired inspiratory and expiratory scans performed on a multislice CT scanner (Siemens Somatom Sensation 64, Erlangen, Germany) using identical technical parameters including submillimeter slice thickness. In order to achieve reproducibility, patients were carefully trained to deeply inhale and hold their breath during each inspiratory scan and to deeply inhale then to forcefully and rapidly exhale and do not breathe for 10 seconds in order to acquire end-expiratory phase images.

Inspiratory scans were evaluated for ground glass opacities (GGOs), nodules, reticulation, honeycombing and airway involvement (including bronchial/bronchiolar wall thickening, bronchiectasis and bronchiolectasis). Bronchial wall thickening was considered present when the ratio between bronchial wall thickness and diameter of bronchus was >0.2 (28, 29). Bronchiectasis was considered present when the broncho-arterial ratio was >1 and bronchiolectasis when peripheral bronchi were visible within 1cm of the costal pleural surfaces (29, 30). CT sections were scored at five levels: 1) the origin of the great vessels, 2) the carina, 3) the pulmonary venous confluence, 4) between levels 3 and 5, and 5) 1cm above the right hemidiaphragm. The following features were quantified at each level: a) The extent of each abnormality described in the previous paragraph was estimated to the nearest 5% for each level and the overall extent was computed as the mean of the five section scores, b) the extend of ILD was graded as follows: 0 : ground-glass opacification alone; 1: fine intralobular fibrosis; 2 : microcystic honeycombing (air spaces up to 4 mm in diameter); 3 : macrocystic honeycombing (air spaces greater than 4 mm in diameter). The five section scores were summed to give the total ILD extent score.

In order to detect air trapping areas on expiratory scans, a side by side comparison of inspiratory and expiratory CT images of the same area was performed and lung attenuation difference was measured by small regions of interest (1–2 cm). Hypodense areas on expiratory scans showing attenuation increase less than 80 HU and involving more than 25% of the lobe, were regarded as air trapping (31, 32). Areas of emphysema were excluded. The extent of air trapping (AT) on expiratory scans corresponding to small airways disease was evaluated by visual assessment using a semiquantitative scoring system estimating the percentage of lung that appeared abnormal on each scan. A 5%-point scoring system, as the one proposed by Webb *et al.* and Stern *et al.* estimated air trapping, on expiratory scans, at three different lung fields for each lung, six lung fields in total for both lungs: upper lungs fields from the lung apices to just above the level of the carina, middle lung fields between the level of the carina and the pulmonary veins and inferior lung fields from the pulmonary veins level till the level of the costophrenic angles (28, 31). At each level and for each lung, a 5-point scale was used to estimate the percentage of air trapping extent visible to each radiologist: 0=no air trapping, 1=1–25% of cross sectional area of the affected lung, 2=26–50%, 3=51–75% and 4=76–100% (28, 31).

HRCT images were independently read by two chest radiologists (E.D., E.M.), blinded to clinical and laboratory data. In cases of discrepancy, images were also evaluated by a third chest radiologist (M.R.) and final decision was then reached by consensus among the three.

Lung function tests

All patients underwent complete PFTs, including spirometry, lung volume and diffusion capacity measurement, at baseline and after one-year treatment. Spirometry, lung volumes using the helium-dilution technique and DLCO (corrected for haemoglobin) using the single breath technique were performed using a computerised system (Jaeger 2.12; MasterLab, Würzburg, Germany). Predicted values were obtained from the standardised lung function testing of the European Coal and Steel Community, Luxembourg (1993). Observed values were expressed as percentage of the predicted value, were compared with individuals of similar sex, age and height and were considered as abnormal if they were <80% of the predicted values adjusted for age, sex, and height (33).

The composite physiologic index (CPI) which represents the extent of fibrosis on HRCT, adjusting for emphysema in patients with idiopathic pulmonary fibrosis (IPF) was calculated using the following formula: 91.0 – (0.65 x percent predicted DLCO) – (0.53 x percent predicted FVC) 1 (0.34 x percentage predicted FEV1) in the RA-ILD group of patients (34).

Laboratory tests

Patients were evaluated at baseline and after one-year treatment for presence of rheumatoid factor (RF) and anti-citrullinated protein antibodies (anti-CCP). Evaluation of RA disease activity included recording of erythrocytes sedimentation rate (ESR) and C-reactive protein (CRP).

Statistics

The Kolmogorov-Smirnov test was used to determine whether the data obtained follow a normal distribution pattern. Group comparisons were made by analysis of variance, Student *t*-test, Wilcoxon rank-sum test, or chi-square testing as appropriate. Linear regression between clinical parameters and the obtained data were analysed with the Linear (Pearson) correlation test and regression analysis was performed for MTX use between groups. Probability values (*p*-values) <0.05 were considered statistically significant. Statistical calculations were performed using SPSS 11.5 software (SPSS, Chicago, IL, USA).

Results

There were no statistically significant differences in demographic parameters between the two patient study groups and the two control groups, respectively (Table I). All patients had moderately to severely active RA. Intraobserver and interobserver reproducibilities were good for the detection and extent of HRCT findings in all pre- and post-treatment scans. HRCT findings are summarised in Table II. PFTs, peripheral blood biomarkers and DAS 28 are presented in Table III. Correlations between HRCT findings and PFTs are analysed in Table IV.

Table I. Demographic data of all patients.

Characteristics	RA-ILD Study Group	RA-non ILD Study Group	RA-ILD Control Group	RA-non ILD Control group	*p*-value
Number of patients	42	40	44	44	
Age	60.05 ± 7.88	62.84 ± 9.52	61.54 ± 8.22	60.84 ± 7.84	p1, p3,p2 NS
Gender (male/female)	15/27	14/26	17/27	18/26	p1, p2, p3 NS
Smoking status (non; current)	30;12	32;8	26;18	38;6	p1, p2, p3 NS
Disease duration (yrs)	8.88 ± 3.37	9.2 ± 3.77	9.12 ± 3.16	9.26 ± 2.34	p1, p2, p3 NS

Values in mean ± SD, Student's *t*-test used for all analyses. NS: non significant, *p*1: RA-ILD *vs*. RA-non ILD study group, *p*2: RA-ILD *vs*. RA-ILD control group; *p*3: RA-non ILD study group *vs*. RA-non ILD control group.

Findings at baseline

In the RA-ILD study group, a usual interstitial pneumonia (UIP) pattern was found in 22 patients (52.5%), non specific interstitial pneumonia (NSIP) in 12 (28.5%) and cryptogenic organising pneumonia (COP) in 8 (19%) (Fig. 1). All 18 patients that showed air trapping areas on expiratory HRCT demonstrated abnormally low $MMEF_{75-25,}$ and abnormally increased RV and RV/TLC values, corresponding to small airways disease. There was no patient with large airways obstruction.

In the RA-ILD control group, a UIP pattern was detected in 13 (29%) patients, NSIP in 16 (35.5%) and a COP pattern in 1 patient (2.2%).

In the RA-non ILD study group, among nodules found, there were two biopsy proven necrobiotic rheumatoid nodules (Table II, Fig. 1, patient 42). On expiratory scans air trapping was depicted in 19 (47.5%) patients. All these patients showed abnormally low maximum mid-expiratory flow ($MMEF_{75-25}$) values at PFTs.

Changes following one year of anti-TNF-α treatment

During the first 6 months of anti-TNF-α treatment 5 patients showed signs and symptoms of lower respiratory tract infection, 3 due to *Streptococcus pneumoniae*, belonging to RA-ILD group, 1 patient due to *Legionella pneumophila* and 1 due to *Listeria monocytogenes*, both belonging to the RA-non ILD study group. Regarding control groups three cases of opportunistic lung infections where depicted, two due to *Pneumocystis Jirovecii* in the RA-ILD control group and one case due to *Mycobacterium Avium intracellulare* in the RA-non ILD control group. In both study and control groups no case of mycobacterium tuberculosis infection was recorded. All the above cases were successfully treated without further complications.

In the RA-ILD study group, overall there was a statistically significant decrease of bronchial wall thickening and of air trapping extent (Table II, Fig. 1, Fig. 2A). There was no significant difference in the extent of ILD pre- and post-treatment (Fig. 3). Patients with decreased air trapping extent showed significantly increased $MMEF_{75-25}$ as well as decreased residual volume (RV) and RV/TLC (total lung capacity) values, *versus* normal range (Table III, Fig. 2B-D). Anti-CCP was found to be significantly decreased (Table III). Correlations between air trapping extent, bronchial wall thickening and PFTs are presented in Table IV.

In the RA-ILD control group, a slight increase in ILD extent score and also in air trapping extent score was recorded, both statistically insignificant (Table II, Fig. 2A). These patients also showed $MMEF_{75-25}$ decrease and RV and RV/TLC increase, although statistically insignificant (Fig. 2B-D). Correlations depicted between air trapping extent, bronchial wall thickening and PFTs are presented in Table IV.

Table II. HRCT findings in RA-ILD, RA-non ILD study groups and in RA-ILD and RA-non ILD control groups at baseline and after 1 year of treatment.

	RA-ILD study group			RA-non ILD study group			RA-ILD control group			RA-non ILD control group		
Characteristics	Baseline	After treatment	*p*-value	Baseline	After treatment	*p*-value	Baseline	After treatment	*p*-value	Baseline	After treatment	*p*-value
n # of patients	42	42		40	40		44	44		44	44	
ILD extent score %	24.52±13.87	24.52±13.43	NS	-	-		15.25±10.42	15.8±11.09	NS	-	-	
Air trapping extent score %	27.36±14.12	17.02±9.63	*p*<0.05	19.00±14.10	16.75±13.86	*p*<0.05	16.56±13.09	16.88±12.06	NS	5.64±6.22	5.96±4.26	NS
Reticular pattern (0;1)	7 ; 35	7 ; 35	NS	40 ; 0	40 ; 0	NS	11 ; 33	11 ; 33	NS	44; 0	44 ; 0	NS
Nodules (0;1)	22 ; 20	22 ; 20	NS	25 ; 15	25 ; 15	NS	30 ;14	29 ; 15	NS	26; 18	26 ; 18	NS
GGOs (0;1)	25 ; 17	26 ; 16	NS	40 ; 0	40 ; 0	NS	28 ; 16	27 ; 17	NS	50 ; 0	50 ; 0	NS
Bronchiectasis (0;1)	17 ; 25	16 ; 24	NS	19 ; 21	19 ; 21	NS	18 ; 26	18 ; 26	NS	17 ; 27	17 ; 27	NS
Bronchiolectasis (0:1)	25 ; 17	25 ; 17	NS	29 ; 11	29 ; 11	NS	24 ; 21	22 ; 22	NS	34 ; 10	33 ; 11	NS
Bronchial wall thickening (0;1)	24 ; 18	28 ; 12	*p*<0.05	17 ; 23	24 ; 16	*p*<0.05	24 ; 20	22 ; 22	NS	20 ; 24	19; 25	NS
Honeycombing (0;1)	20 ; 22	20 ; 22	NS	40 ; 0	40 ; 0	NS	31 ; 13	31 ; 13	NS	44 ; 0	44 ; 0	NS
Air trapping (0;1)	24 ; 18	26 ; 16	NS	21 ; 19	22 ; 18	NS	25 ; 19	24 ; 20	NS	25 ; 25	25 ; 25	NS

Values in mean ± SD. (0;1) is equivalent to (absent ; present). Student's *t*-test used for all analyses. *p*<0.05: statistically significant; NS: non significant; GGOs: ground glass opacities.

Table III. Pulmonary function tests and peripheral blood biomarkers findings in RA-ILD, RA-non ILD groups and in RA-ILD and RA-non ILD control groups at baseline and after 1 year of treatment.

	RA-ILD study group			RA-non ILD study group			RA-ILD control group			RA-non ILD control group		
	Baseline	After treatment	*p*-value	Baseline	After treatment	*p*-value	Baseline	After treatment	*p*-value	Baseline	After treatment	*p*-value
n# of patients	42	42		40	40		44	44		44	44	
FVC % pred	88.12±19.07	88.48±19.93	NS	97.28±16.76	96.07 ±17.00	NS	86.54±16.65	85.18±17.98	NS	98.22±12.53	94.8±15.46	NS
FEV1% pred	87.49±20.98	83.57±20.36	NS	93.00±14.31	90.±16.09	NS	82.53±18.56	79.84±20.11	NS	92.63±13.17	88.20±16.22	NS
FEV1/FVC	82.40±12.48	82.42±10.59	NS	77.99±10.12	75.76±9.29	NS	88.92±9.94	87.52±8.34	NS	80.62±11.05	76.76±8.39	NS
RV % pred	104.2±28.36	91±32.72	*p*<0.05	111.00±32.09	92±24.66	*p*<0.05	111.2±31.67	112.1±30.48	NS	112.4±30.06	113.2±28.61	NS
TLC % pred	77.76±19.75	77.88±21.56	NS	99.46±18.46	100.9±14.30	NS	77.98±17.30	76.93±16.71	NS	97.83±15.69	95.39±16.92	NS
RV/TLC	110.3±17.53	94.0±20.55	*p*<0.05	111.9±19.72	92.1±16.49	*p*<0.05	112.5±20.88	114.6±22.14	NS	113.2±20.34	117.5±23.03	NS
$MMEF_{75/25}$	69.97±25.61	84.89±25.01	*p*<0.05	72.24±22.17	88.90±24.29	*p*<0.05	73.31±17.23	72.56±18.26	NS	74.80±17.95	71.39±19.98	NS
TLCOc SB	79.85±15.93	78.31±20.69	NS	88.33±23.12	88.97±23.98	NS	82.54±18.24	79.56±24.97	NS	83.38±16.15	76.92±22.91	NS
TLCOc/VA	76±21.54	77.4±18.36	NS	98.92±29.99	101.0±27.05	NS	86.44±33.72	84.33±24.22	NS	91.34±28.13	92.04±24.80	NS
DAS 28	4.12±1.29	3.27±1.67	*p*<0.05	4.25±1.50	3.52 ± 1.58	*p*<0.05	4.35 ± 1.3	4.22 ± 1.5	NS	4.4 ± 1.22	4.36 ± 1.24	NS
RF (0;1)	18 ; 24	18 ; 24	NS	18 ; 22	18 ; 22	NS	16 ; 28	16 ; 28	NS	17 ; 27	17 ; 27	NS
Anti-CCP (0;1)	11 ; 31	16 ; 26	*p*<0.05	37 ; 3	37 ; 3	NS	16 ; 28	14; 30	NS	35 ; 9	35 ; 9	NS
ESR	27.48± 14.42	27.96±23.31	NS	23.38±17.99	22.46±17.77	NS	26.46±12.32	29.84±16.22	NS	22.24±12.55	23.55±14.44	NS
CRP	2.76 ± 7.53	2.66±1.31	NS	1.99±0.99	0.45±1.06	NS	2.36±4.53	1.24±1.44	NS	1.44±1.22	0.66±1.22	NS

Values in mean ±SD. (0;1) is equivalent to (absent ; present). Student's *t*-test used for all analyses. $p<0.05$: statistically significant; NS: non significant. FVC: forced vital capacity; FEV1: forced expiratory volume in 1s; RV: residual volume; TLC: total lung capacity; $MEF_{75/25}$: maximum mid-expiratory flow; TLCO: transfer coefficient for carbon monoxide; DAS 28: disease activity score 28; RF: rheumatoid factor; Anti-CCP: anti-citrullinated protein antibodies; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein.

Table IV. Correlations between PFTs parameters, bronchial wall thickening and air trapping in both RA-ILD and RA-non ILD study and control groups at baseline and after 1 year of treatment.

	RA-ILD study group		RA- non ILD study group		RA-ILD control group		RA-non ILD control group	
Correlation	Baseline	After treatment	Baseline	After tr[a]eatment	Baseline	After treatment	Baseline	After treatment
RV *vs*. AT %	r2=0.3135, p=0.0034	r2=0.3135, p=0.0034	r2=0.3236, p=0.0392	r2=0.3132 p=0.052	r2=0.3132, p=0.0033	r2=0.3132, p=0.0033	r2=0.3234, p=0.0392	r2=0.3133 p=0.0534
RV *vs*. $MMEF_{75/25}$	NS	NS	NS	NS	NS	NS	NS	NS
RV/TLC *vs*. AT %	*p*<0.05	*p*<0.05	*p*<0.05	*p*<0.05	*p*<0.05	*p*<0.05	*p*<0.05	*p*<0.05
RV/TLC *vs*. $MMEF_{75/25}$	NS	NS	NS	NS	NS	NS	NS	NS
$MMEF_{75/25}$ *vs*. AT %	-0.324/0.105 *p*=0.0360	-0.324/0.105 *p*=0.0360	-0.35/0.125 *p*=0.0216	-0.35/0.125 *p*=0.0216	-0.395/0.104 *p*<0.05	-0.395/0.104 *p*<0.05	-0.248/0.122 *p*<0.05	-0.248/0.124 *p*<0.05
Bronchial wall thickening *vs*. AT	r2=0.3137, *p*=0.0036	r2=0.3137 *p*=0.0036	r2=0.1920, *p*=0.0059	r2=0.7286, *p*<0.0001	r2=0.3136, *p*=0.0034	r2=0.3136 *p*=0.0034	r2=0.1921, *p*=0.0059	r2=0.7283, *p*<0.0001
Bronchial wall thickening *vs*. AT %	r2=0.2413, *p*=0.0126	r2=0.2356 *p*=0.0139	r2=0.3586 *p*<0.0001	r2=0.3592 *p*<0.0001	r2=0.2413 *p*=0.0126	r2=0.2356 *p*=0.0139	r2=0.3586, *p*<0.0001	r2=0.3592 *p*<0.0001

NS: non significant; AT %: Air trapping extent; AT: presence; $p<0.05$: statistically significant.

In the RA-non ILD study group, there was a a significant decrease of bronchial wall thickening and air trapping extent while there was no significant increase in the extent of inspiratory HRCT findings (Table II, Fig. 1, 2). There was no evidence of new-onset ILD. At post-treatment PFTs, patients with decreased air trapping extent showed also a significant decrease of RV and RV/TLC and increase of $MMEF_{75-25}$ values versus normal range. In the RA-non ILD control group an insignificant increase of air trapping extent score was depicted on expiratory HRCT scans. Concerning PFTs, RV and RV/TLC slightly increased while $MMEF_{75-25}$ values further decreased, all statistically insignificant (Table II, Fig. 2 B-D). No patient of this group developed new-onset ILD.

DAS 28 was found significantly decreased in both RA-ILD and RA-non ILD study groups (Table III). No significant correlation was detected between $MMEF_{75-25}$ values and RV or RV/TLC, respectively, in all groups (Table IV).

Methotrexate regression analysis

In order to establish a clear role of anti-TNF-α agents in this study we performed a detailed regression analysis of the MTX implication in our results. In this view, we tested whether MTX use (dose in mg) altered any of the tested parameters (PFTs, inspiratory HRCT findings and expiratory HRCT findings) between study and control groups. No patient in the study groups and control groups established MTX-induced pneumonitis or exacerbation of previous ILD related to MTX use. Regression analysis demonstrated that no parameter, including HRCT and PFT findings, showed any correlation with MTX

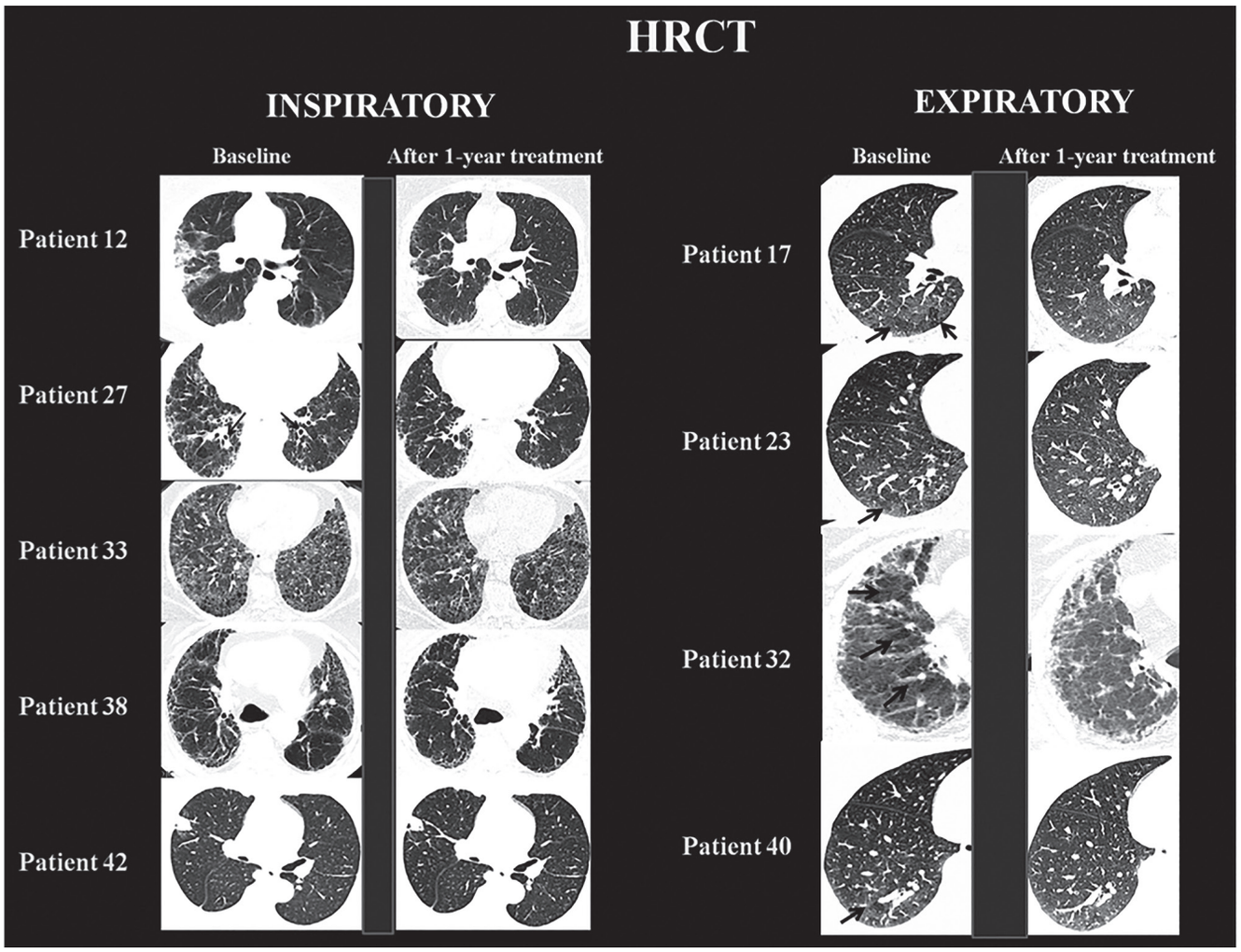

Fig. 1. *Inspiratory HRCT scans* (left): Patient 12 of the RA-ILD group with right lung peripheral consolidations (COP pattern) at baseline and slight decrease of their extent after treatment. Patients 27, 33 and 38 of the RA-ILD group with GGOs, reticulations and honeycombing areas (UIP pattern) at baseline with no significant change of their extent after treatment while bronchial wall thickening improvement was depicted in patient 27 (black arrows). Patient 42 of the RA-non ILD group with a rheumatoid nodule that remained unchanged after treatment.
Expiratory HRCT scans (right): Patients 17 and 23 and 40 of the RA-non ILD group and patient 32 of the RA-ILD group showing substantial improvement of air trapping extent after treatment (black arrows).

use in the RA-non ILD control group, including the subgroup under MTX alone and subgroup under hydroxychloroquinine combined with MTX, as well as between each of the above subgroups and the RA-non ILD study group. These regression analysis results remained unchanged both at baseline and following one year of treatment.

MTX regression analysis revealed a significant negative correlation at one-year study between ILD extent score and MTX dose regarding RA-ILD control (both subgroups, under MTX alone and hydroxychloroquinine with MTX) and RA-ILD study group (p=0.0135, r= -0.3973, r^2= 0.1578). Another negative correlation was also depicted in the RA-ILD study group and the control group with regard to air trapping extent and MTX dose, both at baseline (p=0.0139, r= -0,3908 , r^2= 0.1527) and at one-year follow-up study (p=0.0425, r= -0.3265, r^2= 0.1066), (Fig. 4).

Composite Physiologic Index (CPI)

There were no significant alterations in CPI measurements in the RA-ILD study group before and after anti-TNF-α treatment. Regarding the RA-ILD control group, an increase in CPI was recorded, even though this difference was not statistically significant. CPI positively corellated with honeycombing and GGOs in both RA-ILD study and control groups, at baseline and after one-year treatment. Regarding PFTs, CPI negatively correlated with predicted FVC and TLC in both RA-ILD study and control groups, before and after treatment. CPI ranges. Correlations are recorded in Table V.

Discussion

A statistically significant decrease in the extent of air trapping and bronchial wall thickening was observed after anti-TNF-α treatment in both RA-ILD and RA-non ILD study groups. All patients with decreased air trapping extent on expiratory HRCT showed improvement of $MMEF_{75-25,}$ RV and RV/TLC values versus normal range at post-treatment PFTs. The above suggest that TNF-α

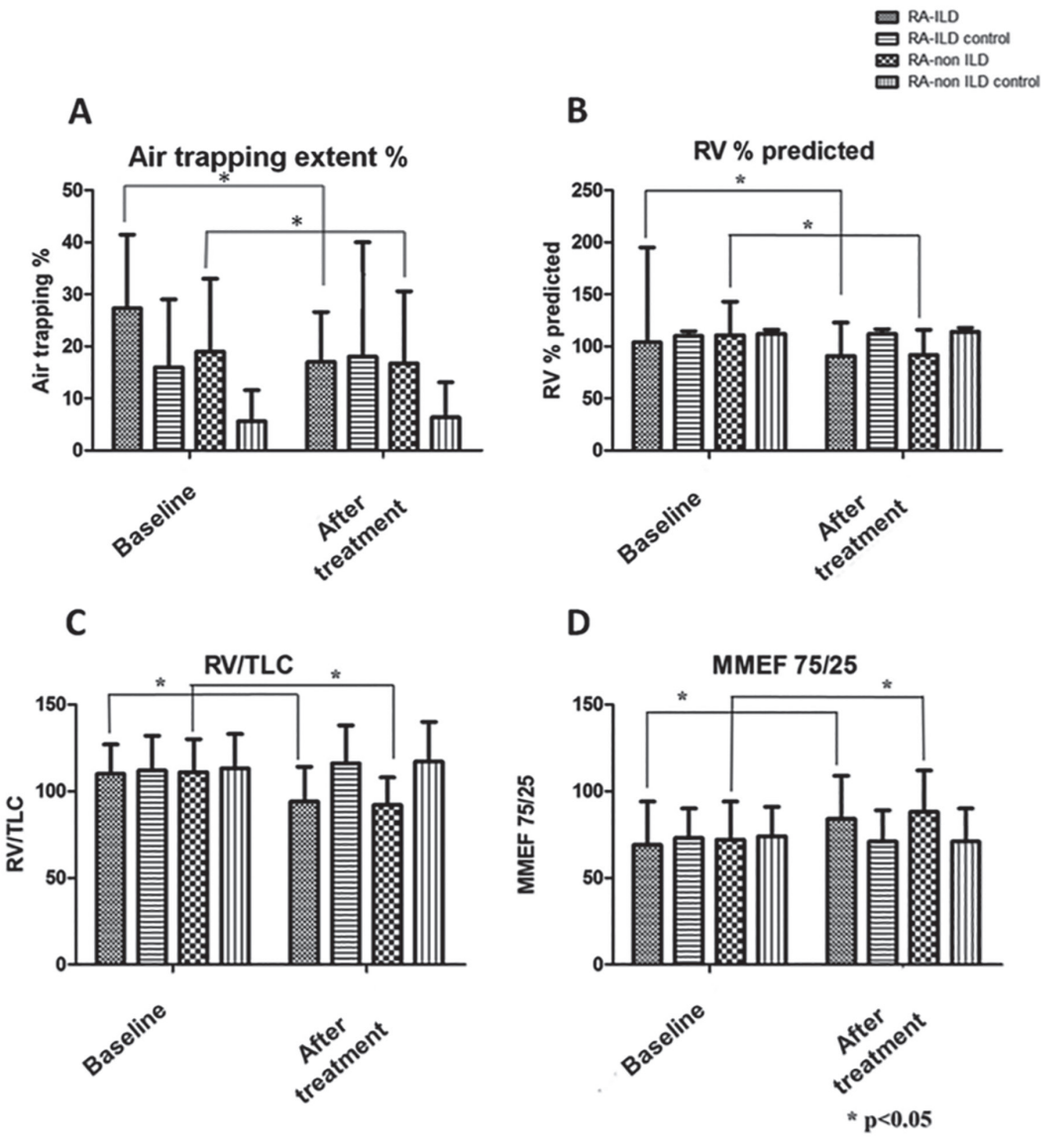

Fig. 2. A) Air trapping extent score significantly decreased after one-year treatment in both RA-ILD and RA-non ILD study groups. A statistically insignificant increase of air trapping extent score was depicted in both control groups after treatment, **B**) and **C**) RV and RV/TLC significantly decreased *versus* normal values after treatment only in RA-ILD and RA-non ILD study groups. In both control groups, RV and RV/TLC insignificantly increased. **D**) $MMEF_{75-25}$ values significantly increased reaching normal range after treatment in both RA-ILD and RA-non ILD study groups. In both control groups $MMEF_{75-25}$ values further decreased, although statistically insignificant.

targeted therapy may play an important role in stabilising and even improving small airways disease in RA patients, either with or without ILD. To the best of our knowledge, the present study, following a case report by Cortot *et al*. is the first prospective study in humans to prove through HRCT findings and PFTs, the beneficial effect of these agents on small airways disease (35).

Our findings can be explained by knowledge of the mechanism of anti TNF-α treatment: TNF-α is a cytokine known to mediate and augment inflammatory reactions and to enhance fibroblast proliferation at the level of the bronchial and bronchiolar wall (36). A study in mice proved that targeted TNF-α overexpression in the lungs is related to chronic inflammatory infiltration of the interstitium by lymphocytes and macrophages, especially localized in areas adjacent to the pleura and bronchioles. TNF-α inhibition has been suggested to reduce inflammation, epithelial loss, fibrosis, and bronchiolar obliteration early in the development of obliterative bronchiolitis (36, 37). Thus, the effect of anti-TNF-α treatment in air trapping extent, bronchial wall thickening and PFTs in our study can be explained by its inhibiting effect on airway wall thickening (36, 38). The increased thickness of the submucosa and proliferation of smooth muscle and connective tissue in RA patients, is related to bronchial wall thickening and air trapping on expiratory HRCT scans (36, 39, 40). This is in accordance with the significant association recorded in the present study between air trapping presence and extent with bronchial wall thickening in all patient groups, at baseline and after treatment.

In the existing literature, the effect of TNF-α in fibrotic disease is controversial. Overexpression of this factor in the lungs has been linked to fibrosis, while inhibition of TNF-α signalling can possibly prevent interstitial lung disease (22, 34). It has been suggested that inflammation leading to recurrent alveolitis may trigger pulmonary fibrosis. It is in this inflammatory stage that anti-TNF-α agents may have a beneficial effect (22).

Moreover, previous studies suggest an association between seropositivity for anti-CCP antibodies, and the presence of ILD in RA patients (11, 41). In ac-

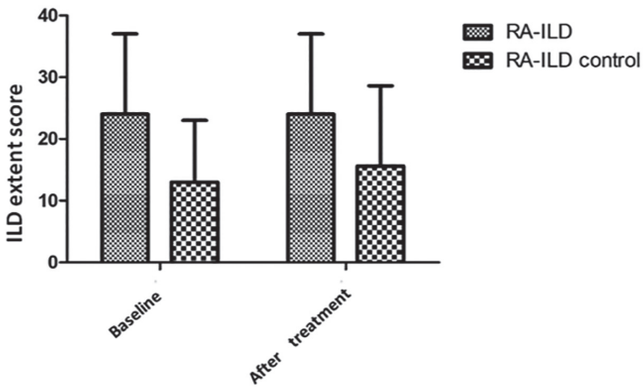

Fig. 3. ILD extent score remained without statistically significant change after anti-TNF-α treatment in RA-ILD study group. In contrast, ILD score insignificantly increased after nb DMARD treatment.

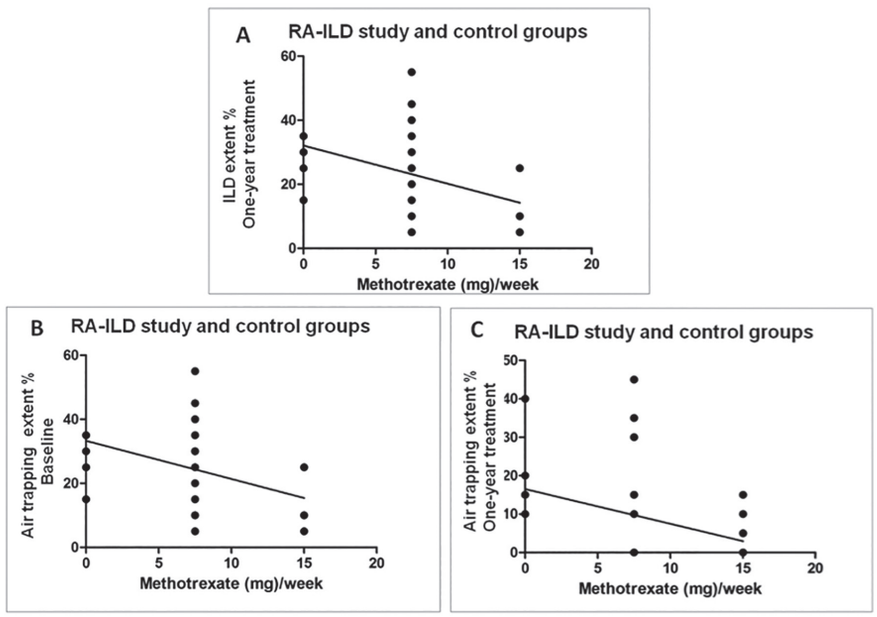

Fig. 4. Methotrexate regression analysis, in both RA-ILD study and control groups revealed, **A**) a significant negative correlation at one-year treatment study between ILD extent score and MTX dose (p=0.0135, r=-0.3973, r^2=0.1578). Another negative correlation was also depicted between air trapping extent and MTX dose, in RA-ILD control and study groups, both, **B**) at baseline (p=0.0139, r=-0.3908, r^2=0.1527) and C) at one-year treatment study **C**) (p=0.0425, r=-0.3265, r^2=0.1066).

cordance, we detected a statistically significant higher serum level of anti-CCP antibodies in the RA-ILD study group compared to the RA-non ILD. Following treatment, serum levels of anti-CCP significantly decreased in the RA-ILD study group while such decrease was not registered in the RA-ILD control group. In the present study, most patients in the RA-ILD study group, according to PFT parameters and peripheral blood biomarkers, appeared in an inflammatory stage before anti-TNF-α treatment and this is probably the reason why anti-TNF-α appeared to stabilise inspiratory HRCT findings and reduce anti-CCP antibodies levels in this group. More trials are needed to investigate the effect of anti-TNF-α treatment in RA patients at a non-inflammatory stage.

There are several reports suggesting that anti-TNF-α agents may trigger pulmonary fibrosis in RA patients (20-22). During this one-year follow-up no patient developed exacerbation or progression of pre-existing ILD while none of the RA-non ILD group patients established new-onset ILD. All the above, in accordance with other studies, support the safety of anti-TNF-α agents in patients with RA-ILD with regard to the occurrence and extent of ILD (23-26, 42).

Methotrexate-induced lung toxicity has been widely described in literature (43-46). Pneumonitis related to MTX appears to occur less often than previously thought (47). In our study, no patient developed MTX-induced pneumonitis or any other type of lung toxicity related to MTX supporting the reported favourable safety of its use (48). Furthermore, no significant correlation was recorded between MTX use, PFTs and HRCT findings extent in the RA-non ILD study group and control groups. In contrast, in the RA-ILD study group and the control group, methotrexate regression analysis showed a significant negative correlation at one-year interval between ILD extent score and MTX dose further supporting the idea that low dose MTX treatment may be related to more extended HRCT findings of MTX-induced ILD comparing to higher dose treatment (49, 50). Another negative correlation was also depicted in the RA-ILD study group and the control group with regard to air trapping extent and MTX dose, both at baseline and at one-year follow-up study. These negative correlations were not followed by similar negative correlations regarding PFT parameters. Therefore, even though the above correlations may indicate a possible relation between low dose MTX treatment and a more extended MTX-induced pneumonitis as well as low dose MTX and more extended air trapping on expiratory HRCT, larger scale and longer term studies are needed to draw safer conclusions.

The CPI is an important diagnostic tool that strongly correlates with the CT extent of pulmonary fibrosis and is linked to mortality in histologically proven UIP and idiopathic pulmonary fibrosis (IPF) (33). In our study, CPI levels did not differ significantly after treatment in all groups, thus supporting the stabilising role of treatment with regard to lung fibrosis. It is not a surprise that the CPI correlated positively with honeycombing and GGOs on inspiratory HRCT scans and negatively with predicted FVC and TLC, since all these parameters are linked to restrictive lung disease.

The risk of opportunistic infections associated with anti-TNF-α treatment is widely known (51). During the first 6 months of anti-TNF-α treatment only 5 patients showed signs and symptoms

Table V. CPI variations and correlations with honeycombing, GGOs, FVC and TLC in both RA-ILD study and control group at baseline and after 1-year treatment.

	RA-ILD study group		RA-ILD control group	
	Baseline	After treatment	Baseline	After treatment
CPI range	68.32+6.64	66.78+6.83 *p*1= NS	70.34+9.48	72.56+10.45 *p*2=NS
CPI *vs*. Honeycombing	r=+0.6270 r2=0.3931 *p*=0.0008	r=+0.5324 r2=0.2835 *p*=0.0061	r=+0.6334 r2=0.3982 *p*=0.0008	r=+0.5282 r2=0.2893 *p*=0.0062
CPI *vs*. GGOs	r=+0.4002 r2=0.1602 *p*=0.0474	r=+0.3906 r2=0.1503 *p*=0.0544	r=+0.4232 r2=0.2834 *p*=0.0481	r=+0.4133 r2=0.2832 *p*=0.0543
CPI *vs*. FVC	r=-0.4394 r2=0.1931 *p*=0.0280	r=-0.4293 r2=0.1843 *p*=0.0322	r=-0.4394 r2=0.1931 *p*=0.0280	r=-0.4286 r2=0.1897 *p*=0.0290
CPI *vs*. TLC	r=-0.9800 r2=0.9605 *p*<0.0001	r=-0.9574 r2=0.9167 *p*<0.0001	r=-0.9678 r2=0.9571 *p*<0.0001	r=-0.9623 r2=0.9142 *p*<0.0001

CPI: Composite Physiologic Index; *p*1: RA-ILD study group CPI at baseline *vs*. post-treatment; *p*2: RA-ILD control group CPI at baseline *vs*. post-treatment; GGOs: ground glass opacities; NS: non significant; $p<0.05$: statistically significant.

related to opportunistic lower respiratory tract infection. Patients of both study groups were successfully treated without complications while no mycobacterium tuberculosis or atypical mycobacterial infection was recorded in both study groups.

Limitations of this study include the relatively small number of patients, the wide age range and duration of disease as well as the relatively short term of follow-up. Furthermore, most patients in the RA-ILD and RA-non ILD study groups were treated with infliximab, while only a small percentage received other anti-TNF-α agents which did not permit further statistic correlations to be made for each agent separately. Even though a one-year follow-up period is adequate to detect any significant short or medium term effect regarding pulmonary manifestations, more long-term studies are needed in order to solidify the safety of these agents. In addition, other biomarkers such as KL-6 reported in patients with ILD, were not investigated (21).

In conclusion, the results of the present study support the beneficial effect of anti-TNF-α agents with regard to small airway disease by improving air trapping extent, bronchial wall thickening and $MMEF_{75-25}$, RV and RV/TLC values. Anti-TNF-α agents were not associated with ILD development or progression and opportunistic infections were limited, thus supporting the safety profile of this treatment. In patients with RA-ILD anti-TNF-α treatment had a stabilising effect with regard to ILD, probably more efficiently during the inflammatory stage of disease.

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