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ORIGINAL ARTICLE

Drug levels, anti-drug antibodies, and clinical efficacy of the anti-TNFα biologics in rheumatic diseases

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Abstract The objectives of this study are to evaluate the effect of anti-drug antibodies on the clinical efficacy and withdrawal rate of the anti-TNFα biologics in patients with rheumatic diseases. Consecutive patients with rheumatic diseases recently commenced on anti-TNFα biologics were recruited. Serum samples were collected for assay of drug level and antibody titer against the corresponding biologics. Comparison of the clinical efficacy and drug retention rate was performed between patients with and without anti-drug antibodies. Fifty-eight Chinese patients were studied (64 % women; age 47.8±12.9 years; disease duration 6.7±6.4 years). The proportion of patients using infliximab (IFX), adalimumab (ADA), and etanercept (ETN) was 41, 28, and 31 %, respectively. Antibodies against IFX, ADA, and ETN were demonstrated in 12(50 %), 5(31 %) and 0(0 %) patients, respectively. Patients who developed anti-drug antibodies had significantly lower levels of the corresponding drugs (IFX level: 0.004±0.01 vs 3.81±3.49 μg/ml; $p=0.002$; ADA level: 0.0 vs 7.6±8.3 μg/ml; $p=0.008$). Anti-drug antibody-positive patients had a significantly higher cumulative drug withdrawal rate due to inefficacy (64.7 and 71.8 % vs 10.3 and 10.3 % at month 12 and month 24, respectively; $p<0.001$). In rheumatoid arthritis and psoriatic arthritis, non-responders was significantly more frequent in antibody-positive patients (54 vs 13 %; $p=0.01$). In spondyloarthritis, the improvement in ankylosing spondylitis disease activity score was significant in patients without antibodies (3.89±0.82 to 2.22±0.86; $p=0.01$) but not in those with anti-drug antibodies (3.40±1.67 to 3.23±1.40; $p=0.73$). We concluded that the presence of neutralizing antibodies is associated with lower serum levels of the anti-TNFα biologics, leading to lower efficacy and higher withdrawal rate.

Keywords Antibodies · Biologics · Efficacy · Immunogenicity · Neutralizing · Tolerance

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Introduction

The anti-TNFα biologics are powerful agents for the treatment of various rheumatic and inflammatory disorders such as rheumatoid arthritis (RA), spondyloarthritis (SpA), psoriatic arthritis (PSA), and inflammatory bowel diseases. Their efficacy in these diseases has been demonstrated in large double-blind randomized placebo-controlled trials [1]. In patients with RA, anti-TNFα therapy has proven benefits in both early and established disease [2, 3].

Despite the availability of the anti-TNFα agents for the treatment of chronic inflammatory arthritides, a substantial proportion of patients do not respond or only respond partially to treatment (primary failure). Some patients have an initial good response but the efficacy of some of these agents does not persist (secondary failure). One of the known reasons for secondary failure of these anti-TNFα agents is their immunogenicity. Both chimeric and human monoclonal antibodies can induce neutralizing antibodies, which are known as human anti-chimeric antibodies (HACA) and human anti-human antibodies, respectively [4]. The proportion of patients with various rheumatic diseases who develop neutralizing antibodies to the monoclonal anti-TNFα biologics such as infliximab (IFX) and adalimumab (ADA) ranges from 8-75 % [5]. This wide range of figures is due

to many factors that include technical differences in the detection of the neutralizing antibodies, standardization of the assays, duration of treatment of the biologics, underlying inflammatory diseases and whether concomitant immunosuppressive agents such as methotrexate (MTX) and azathioprine were administered or not.

It is recognized that the clinical efficacy of the anti-TNFα agents is undermined by the presence of neutralizing antibodies, which is associated with lower serum drug levels [6, 7]. In a 6-month prospective study of patients with RA treated with either IFX or ADA, the response at 6 months was strongly correlated with the levels of IFX/ADA and the presence of antibodies against these two biologics [8]. More recent cohort studies have also demonstrated that the presence of anti-ADA antibodies is associated with a reduced clinical efficacy and higher rate of treatment failure of ADA in the treatment of RA, both in short-term (28 weeks) [9] and on long-term follow-up (3 years) [10].

There have not been any studies of neutralizing antibodies in Chinese patients receiving the anti-TNFα biologics. The current study is undertaken to evaluate the incidence of immunogenicity of the anti-TNFα agents in a group of Chinese patients receiving these therapies, and its effect on the clinical efficacy and drug retention rate.

Patients and methods

Study population and study logistics

Between December 2011 and February 2012, consecutive patients with RA, SpA, PSA or other rheumatic diseases that were treated with the anti-TNF agents in the Day Care Center of Pok Oi Hospital, Hong Kong were invited to participate in this study. The inclusion criteria were: (1) patients aged ≥18 years; and (2) patients commenced on any one of the anti-TNFα agents for the first time (new users) for their active disease for less than 24 months. Patients who did not fulfill the inclusion criteria were excluded from this study. Written consent was obtained from all the participants and the study was approved by the Research and Ethics Committee of our hospital.

Blood samples were collected from patients in the morning just before they received a new intravenous or subcutaneous dose of the anti-TNFα agents. These samples were assayed for the following: (1) levels of antibodies to these anti-TNFα agents; and (2) corresponding serum levels of the anti-TNFα agents.

Analyses will be carried out on the correlation between the presence and absence of neutralizing antibodies and drug levels on the observed clinical efficacy in terms of improvement/change in disease activity indices since the baseline visit and the drug retention rate over time.

Assessment of disease activity

The disease activity of RA and PSA in this study was assessed by the disease activity score using the 28-joint count system (DAS28) (swollen joint count, tender joint count, ESR and general health) as described elsewhere [11]. The disease activity of SpA was assessed using the Bath ankylosing spondylitis (AS) disease activity index (BASDAI) (on a 100-mm visual analog scale) and the AS disease activity score (ASDAS) (using C reactive protein (CRP)) as described previously [12, 13].

Dosage of the anti-TNFα agents used

All patients in this study received government subsidy for the anti-TNFα biologics and followed our pre-defined indication for these agents. The criteria for use of anti-TNFα treatment in RA patients were: (1) DAS28 score≥5.1; and (2) failed ≥2 conventional disease modifying anti-rheumatic drugs (DMARDs). For PSA patients, the criteria were: (1) ≥4 swollen joints; (2) ≥4 tender joints; (3) raised ESR or CRP; and (4) failed ≥3 conventional DMARDs. For axial SpA patients, the criteria were: (1) BSADAI≥4; (2) raised ESR or CRP, or imaging evidence of sacroiliitis; and (3) failed ≥3 non-steroidal anti-inflammatory drugs (NSAIDs). For patients with peripheral SpA, the criteria were: (1) ≥4 swollen joints; (2) ≥4 tender joints; (3) raised ESR or CRP; and (4) failed ≥2 conventional DMARDs.

The dosage of IFX used in our patients was 3 mg/kg per dose for RA (augmented to 5 mg/kg when response was suboptimal at week 16), 5 mg/kg per dose for SpA and PSA, given as intravenous infusion at baseline, week 2, 6, and then every 6–8 weeks. Etanercept (ETN) was given subcutaneously at 50 mg per dose every week. ADA was given subcutaneously at 40 mg per dose every 2 weeks.

Definition of clinical response and criteria for drug withdrawal

The EULAR response criteria were used to assess the clinical response of our RA and PSA patients to anti-TNFα treatment compared to baseline [14]. Briefly, the criteria were: good response—an improvement of >1.2 and a present score of ≤3.2; moderate response—an improvement of >0.6 to ≤1.2 and a present score of ≤5.1, or an improvement of >1.2 and a present score of >3.2; no response—an improvement of ≤0.6, or patients with an improvement of >0.6 to ≤1.2 and a present score of >5.1. For patients with SpA, improvement was defined as a reduction of BASDAI score of ≥50% and an absolute reduction of ≥20 points.

The criteria for discontinuation of anti-TNFα therapy in RA and PSA patients due to inefficacy were: RA—failure of improvement of the DAS28 score by 1.2 points after

16 weeks; PSA—failure of improvement of the tender and swollen joint counts by 30 %, failure of improvement of physicians' global assessment by 20 (on a 0–100-mm visual analog scale), or failure of improvement of ESR/CRP after 16 weeks. For patients with SpA, when improvement of BASDAI was less than 50 % or absolute improvement <20 after 16 weeks, anti-TNFα treatment would be stopped. Patients would then be assessed for the use of another anti-TNFα agent or the non-TNFα biologics (for RA). For statistical analyses, for patients who were discontinued the first anti-TNFα agent, the time to event was counted from baseline to the time of treatment cessation.

Assay of drug and anti-drug levels

Drug levels were measured at Sanquin using in-house-developed ELISAs. Maxisorp ELISA plates were coated overnight with 2 μg/ml monoclonal anti-TNF-7 (Sanquin) in Phosphate buffered saline (PBS) at room temperature (RT) for infliximab and adalimumab, or with 2 μg/ml monoclonal anti-TNF-7 (Sanquin) in Phosphate buffered saline (PBS) at RT for etanercept. After five times washing with PBS/0.02 % Tween (PT), plates were incubated for 1 h at RT with recombinant TNFα (0.01 μg/ml for infliximab and adalimumab; 0.05 μg/ml for etanercept) (Strathmann Biotech HmbH, Hannover, Germany) diluted in high-performance ELISA buffer (HPE, Business Unit reagents, Sanquin). Next, the plates were washed and incubated for 1 h with patient serum which was serially diluted in HPE. Subsequently, the plates were washed with PT and incubated for 1 h with either biotinylated infliximab (0.25 μg/ml), biotinylated adalimumab (0.125 μg/ml) or biotinylated etanercept (1 μg/ml) specific rabbit anti-idiotype antibody in HPE. After washing, streptavidin-poly-HRP (Sanquin) (40 ng/ml for infliximab and adalimumab, 80 ng/ml for etanercept) in HPE was added for 1 h at 37 °C. After washing the ELISA was developed with 100 μg/ml tetramethylbenzidine in 0.11 M sodium acetate (pH 5.5) containing 0.003 % (*v*/*v*) H_2O_2. The reaction was stopped with 2 M H2SO4. Absorption at 450 nm was measured with an ELX808IU reader (Bio-tek Instruments Inc., USA). Results were related to a titration curve of infliximab on each plate. The lowest level of quantification was 0.002 mg/l.

To measure anti-drug-antibodies, an in-house-developed RIA was used. For infliximab and adalimumab, 1 μl of serum diluted in Freeze buffer was incubated with 1 mg protein A Sepharose (GE healthcare, Chalfont St. Giles, UK) in 800 μl of total volume. After overnight incubation, samples were washed and 125I radioactively labeled infliximab or adalimumab F(ab′)2 fragments were added. After overnight incubation, unbound radiolabel was washed out and Sepharose-bound radioactivity was measured. For etanercept, 50 μl of serum was incubated with etanercept-coupled Sepharose in a total volume of 800 μl. After overnight incubation, samples were washed and ^{125}I-radioactively labeled etanercept was added. After overnight incubation, unbound radiolabel was washed out and Sepharose-bound radioactivity was measured. Results of these tests are commonly expressed by Sanquin as arbitrary units per ml, where 1 AU/ml equals approximately 10μg/L. The lower limit of quantification is 12 AU/ml.

Statistical analyses

Unless otherwise stated, values in this study were expressed as mean±standard deviation. Comparison of continuous variables between two groups was performed by the non-parametric Mann–Whitney rank sum test. Categorical variables were compared by the Chi-square test. When the frequency was <5, the Fisher's exact test was used. The disease activity scores of various rheumatic diseases at the time of blood collection and baseline were compared by the Wilcoxon's matched pair test.

The cumulative probability of withdrawal of the anti-TNF agent over time due to inefficacy was studied by the Kaplan–Meier's method. Time zero was the time of commencement of the biological agents and data were censored at the time of the last dose of administration of the drugs. The cumulative drug retention rate in patients with and without anti-drug antibodies was compared by the log rank test. The hazard ratio and 95 % confidence interval for drug withdrawal in patients with and without the anti-drug antibodies was computed by the Cox regression method.

Statistical significance was defined as a two-tailed *p* value of <0.05. All statistical analyses were performed using the SPSS program, version 11.5 for Windows Vista.

Results

Characteristics of the study population

Sixty patients were invited but two declined to participate in this study. Finally, 58 patients were recruited (64 % women). The mean age at the time of blood collection was 47.8±12.9 years and the duration of the underlying diseases was 6.7±6.4 years. The underlying diseases of these patients that required anti-TNFα treatment were: RA (*N*=35; 60 %), SpA (*N*=12; 21 %), PSA (*N*=10; 17 %) and refractory idiopathic uveitis (2 %). The proportion of patients treated with IFX, ADA, and ETN, was 41, 28, and 31 %, respectively. The mean duration of therapy at the time of blood collection was 11.8±8.0, 9.3±4.0, and 13.3±7.7 months for IFX, ADA, and ETN users, respectively.

Table 1 shows the baseline clinical characteristics and disease activity scores of our patients. At the time of anti-

TNFα treatment, MTX, alone or combined with other DMARDs, was used in 29 (83 %) RA and 8 (80 %) PSA patients, respectively. Seven (58 %) SpA patients received sulfasalazine but none were treated with MTX.

Antibodies to the anti-TNFα agents and drug levels

Antibodies against IFX, ADA and ETN were demonstrated in 12 (50 %), 5 (31 %), and 0 (0 %) patients, respectively ($p=0.002$). Patients who developed anti-drug antibodies had significantly lower levels of the corresponding drugs (IFX level: 0.004±0.01 vs 3.81±3.49 μg/ml; $p=0.002$; ADA level: 0.0 vs 7.6±8.3 μg/ml; $p=0.008$). Similar findings were observed for current users of IFX (drug level: antibody-positive 0.01±0.02 vs antibody-negative 5.08±3.1 μg/ml). All current users of ADA were negative for anti-ADA antibodies.

Relationship between anti-drug antibody and clinical efficacy

In patients with RA and PSA, treatment with the anti-TNFα agents was associated with a statistically significant change in the mean DAS28 scores compared to the baseline. However, the magnitude of change in DAS28 scores in antibody-positive patients was smaller than that of the antibody-negative comparators (from 6.15±1.39 to 4.94±1.54 vs from 5.51±1.18 to 3.73±1.26). This difference in DAS28 improvement was clinically relevant because the proportion of non-responders (EULAR RA response criteria) after anti-TNFα treatment was significantly higher in patients with anti-drug antibodies than those without (54 vs 13 %; $p=0.01$) (Table 2). Table 3 shows the clinical response of these patients according to the anti-TNFα agents used. The rate of treatment failure (no response) was lower in users of ETN (8 %) compared to the other two TNFα inhibitors (30 % in IFX and 33 % in ADA).

In patients with SpA, the mean improvement in ASDAS score was significant in patients without antibodies (3.89±0.82 to 2.22±0.86; $p=0.01$) but not in those with anti-drug antibodies (3.40±1.67 to 3.23±1.40; $p=0.73$) (Fig. 1). Similar findings were demonstrated with the BASDAI scores—reduction of BASDAI scores in antibody-positive patients ($N=3$) was not significant (48.5±26 to 44.5±22; $p=0.80$) but the improvement in BASDAI was statistically significant in patients without anti-drug antibodies ($N=9$) (63.2±25 to 33.9±22; $p=0.03$). None of the patients with anti-drug antibodies met our improvement criteria, compared to 44 % of the antibody-negative patients met the criteria. Regarding the clinical efficacy of individual anti-TNFα agent, the only patient treated with IFX (anti-drug antibody-negative) met our improvement criteria. Three of five (60 %) and two of six (33 %) patients treated with ETN and ADA, respectively, showed clinical improvement.

The remaining patient had idiopathic uveitis that was refractory to conventional immunosuppressive agents. He was treated with ADA but there was no clinical response. He was tested positive for the anti-ADA antibody.

Relationship between anti-drug antibody and drug retention rate

Seventeen patients had discontinuation of anti-TNFα therapy at the time of analysis. Clinical inefficacy was the reason in 16 patients while the remaining patient suffered from allergic skin lesions to IFX. Of the 17 patients with anti-drug antibodies, 12 (71 %) had withdrawal of anti-TNFα therapy due to inefficacy, which was significantly more frequent than those without antibodies (4/41 or 10 %)

Table 1 Clinical characteristics of the patients at the time of anti-TNFα treatment

	RA ($N=35$)	PSA ($N=10$)	SpA ($N=12$)
	Mean±SD; *N* (%)		
Age, years	50.3±12.4	49.3±12.4	40.9±12.2
Female sex	26 (74)	7 (70)	4 (33)
Disease duration, years	5.6±5.5	5.0±2.9	11.4±8.8
Concomitant DMARDs			
MTX	29 (83)	8 (80)	0 (0)
MTX dosage	10.4±5.9	11.8±9.0	0 (0)
SSz	8 (23)	1 (10)	7 (58)
HCQ	12 (34)	0 (0)	0 (0)
LEF	2 (6)	0 (0)	0 (0)
DAS28 score	5.62±1.44	5.42±1.23	–
BASDAI	–	–	58.8±24.7
ASDAS	–	–	3.72±1.08

TNF tumor necrosis factor, *RA* rheumatoid arthritis, *PSA* psoriatic arthritis, *SpA* spondyloarthritis *SD* standard deviation, *DMARDs* disease modifying anti-rheumatic drugs, *MTX* methotrexate, *SSz* sulfasalazine, *HCQ* hydroxychloroquine, *LEF* leflunomide, *DAS28* disease activity score using the 28-joint counts, *BASDAI* Bath ankylosing spondylitis disease activity index, *ASDAS* ankylosing spondylitis disease activity score

Table 2 EULAR response after anti-TNFα treatment in RA and PSA patients

EULAR response	Anti-drug positive (%)	Anti-drug negative (%)
Good	2 (15)	9 (28)
Moderate	4 (31)	19 (59)
None	7 (54)	4 (13)
Total	13 (100)	32 (100)

$p=0.01$

Table 3 EULAR response after anti-TNFα treatment in RA and PSA patients

EULAR response	IFX (%)	ETN (%)	ADA (%)
Good	4 (17)	3 (23)	4 (44)
Moderate	12 (52)	9 (69)	2 (22)
None	7 (30)	1 (8)	3 (33)
Total	23 (100)	13 (100)	9 (100)

($p<0.001$). Compared to those without anti-drug antibodies, antibody-positive patients had a significantly higher cumulative drug withdrawal rate due to inefficacy (64.7 and 71.8 % vs 10.3 and 10.3 % at month 12 and month 24, respectively; $p<0.001$) (Fig. 2).

Factors associated with the development of anti-drug antibodies

Logistic regression was performed for factors that were associated with the development of anti-drug antibodies to either IFX or ADA (ETN-treated patients were excluded as none of them developed antibodies to the drug). As all the SpA patients were not treated with MTX, multi-collinearity test did not suggest putting the underlying rheumatic diagnosis as a covariate in the regression model. As shown in Table 4, the only factor that was associated with development of antibodies to the monoclonal anti-TNFα agents was female sex (OR 8.3 [1.002–69]; $p<0.05$). Concomitant use of MTX was protective but its effect was not statistically significant (OR 0.24 [0.04–1.61; $p=0.14$].

Discussion

Immunogenicity refers to the development of antibody response to exogenous/foreign agents such as vaccines and drugs. While immunogenicity is the desired effect for vaccine administration, the development of neutralizing antibodies to therapeutic drugs may greatly alter their pharmacokinetics, leading to reduced half life and efficacy. In the past decade, the anti-TNF agents have become standard therapies for patients with inflammatory arthritides who respond inadequately to conventional DMARDs [2, 3]. The development of neutralizing antibodies to the anti-TNFα monoclonal antibodies during the course of treatment is a well recognized phenomenon, regardless of whether they are chimeric and fully humanized compounds [15].

IFX is a mouse-human chimeric IgG1 monoclonal antibody that targets both transmembranous and soluble TNFα. In monkey experiments, the administration of anti-IFX IgG following one dose of intravenous IFX shortened the mean terminal serum half life of the drug from 5 days to 10 h [16]. Clinical studies in RA patients also demonstrated that the presence of anti-IFX antibodies reduced the serum concentration of the corresponding anti-TNFα monoclonal antibodies, leading to reduced efficacy [6–8, 17, 18]. The prevalence of anti-drug antibodies in IFX users ranges from 19 to 50 %

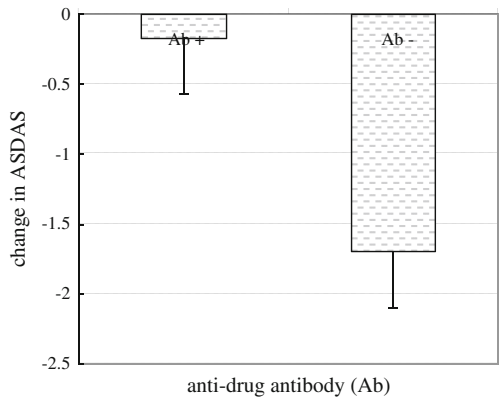

Fig. 1 Change in AS DAS in patients with and without anti-drug antibodies

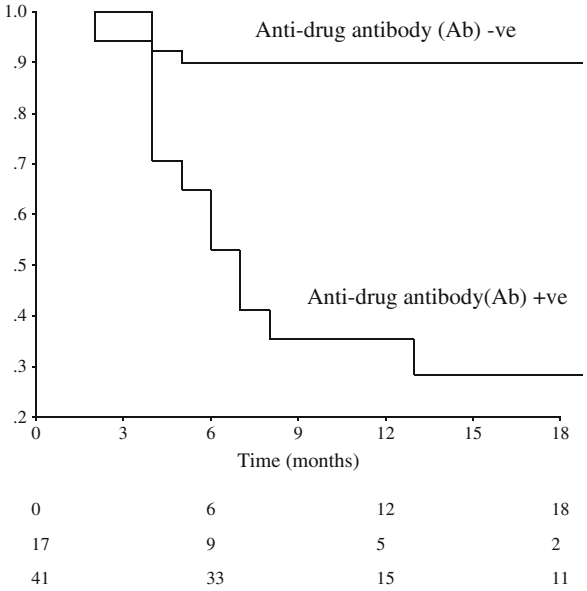

Fig. 2 Cumulative probability of drug retention with regard to the presence or absence of anti-drug antibodies

Table 4 Factors associated with anti-drug antibody development in infliximab and adalimumab users

Covariates	Odds ratio (95 % confidence interval)	*p*
Female sex	8.3 (1.002–69)	0.049
Age, per year	0.95 (0.89–1.01)	0.10
Disease duration, per year	0.96 (0.86–1.08)	0.52
Concomitant methotrexate	0.24 (0.04–1.61)	0.14

depending on the assay method, timing of blood collection (i.e., duration of IFX use) and the underlying disease [6–8, 17, 18]. This is consistent with this study in which 50 % of our IFX users developed antibodies against the drug after administration for a mean of 11.8 months.

Although ADA is a fully humanized anti-TNFα monoclonal antibody, neutralizing antibodies were still reported in 17–28 % of users [8–10]. The 31 % prevalence of anti-ADA antibodies in our ADA users is similar to the experience in other ethnic groups. On the contrary, ETN is a fusion protein consisting of human TNF receptor-2 and the Fc portion of human IgG1. Antibodies to ETN may develop against the fusion region but they do not affect the binding of the molecule to TNF. Several clinical trials have reported anti-ETN antibodies in 3–6 % of patients but in all cases, the antibodies were non-neutralizing and did not appear to affect its efficacy and safety [19, 20]. In two other cohort studies, anti-ETN antibodies could not be detected [21, 22]. The absence of antibodies to ETN in our study confirmed the above observation.

Patients with antibodies to one anti-TNFα monoclonal antibody are more likely to develop antibodies to another anti-TNFα. In a study of 52 RA patients who switched from IFX to ADA because of inefficacy, anti-ADA antibodies developed in 33 % of patients who were previously tested positive for the anti-IFX antibodies, as compared to 16 % in patients without the anti-IFX antibodies and 18 % in 183 ADA users who were never treated with IFX before [23]. Interestingly, those patients who were anti-IFX antibody-positive had better clinical response (improvement in DAS28 scores) after 28 weeks' treatment with ADA than those without the anti-IFX antibodies. This illustrates that if the treatment failure to IFX is due to the occurrence of neutralizing antibodies, the chance of having a clinical response is higher when shifting to another anti-TNFα agent, whereas when treatment failure to IFX is not due to anti-drug antibodies, patients are less likely to respond to another anti-TNFα with similar mechanism of action. Similar findings were observed in 89 RA patients who were shifted from either IFX to ETN [24]. Those who were tested positive for anti-IFX or anti-ADA antibodies were more likely to respond to ETN than those without anti-drug antibodies.

In addition to the molecular structures of the anti-TNFα agents themselves, other factors have been reported to influence the development of immunogenicity [25]. Concomitant use of MTX with either ADA or IFX in RA patients was well recognized to reduce the incidence of anti-drug antibody [10, 26], which partly explained the synergistic effects of MTX and the anti-TNFα agents. Patients with more active RA, erosive disease and longer disease duration at baseline were more likely to develop anti-ADA upon ADA treatment [10]. The underlying disease that required biological therapy may also influence the incidence of anti-drug antibodies. For instance, in patients with systemic lupus erythematosus treated with rituximab, up to two thirds of patients developed the HACA response to this chimeric monoclonal antibody [27]. In our study, we demonstrated that female patients were more likely to generate anti-drug antibodies. This is consistent with the general knowledge that women are more prone to autoimmunity. Although concomitant MTX use was protective against the development of anti-drug antibodies in our patients, its effect did not reach statistical significance, probably related to the limited sample size of our study.

Immunogenicity of the anti-TNFα agents has also been linked to adverse effects such as infusion reaction. From the experience of IFX treatment of Crohn's disease, it was reported that those patients who were positive for anti-IFX antibodies had a higher relative risk (2.4) of developing infusion reaction to the drug [28]. Patients with infusion reaction had significantly lower levels of IFX than those without this reaction. A postulated mechanism of infusion reaction was the formation of drug-anti-drug immune complexes which activated the complement pathway [28]. More recently, anti-ADA antibodies were also linked to an increase of the risk of arterial and venous thrombosis in RA patients treated with the drug by six- to sevenfold [29]. However, the absolute incidence of this complication was very low in this study and this observation requires confirmation in larger cohorts with longer period of follow-up.

There are some limitations of our study. First, the sample size is not large enough to allow analysis of the factors affecting immunogenicity of the anti-TNFα biologics according to different underlying diseases and individual conventional DMARDs. Second, the assay of the drug levels and anti-drug antibodies was performed at a single time point. The effect of the neutralizing antibodies on long-term prognosis of rheumatic diseases such as joint damage and disability was not the focus of the present work. Nevertheless, we concluded that the development of neutralizing antibodies to the anti-TNFα monoclonals was associated with reduction in serum levels of the agents, leading to reduction in clinical efficacy and drug retention rate, and that female patients were more prone to immunogenicity of the anti-TNFα monoclonals. A longer term prospective study of the drug retention rate, clinical efficacy and outcome in a larger sample of patients with rheumatic diseases treated with the anti-TNFα agents with regard to the anti-drug antibodies is underway in Hong Kong.

Disclosures This is an investigator-initiated study. The cost of the assay of drug level and anti-drug antibodies was sponsored by a research grant from Pfizer. Pfizer was not involved in the study design, data collection or analyses. The authors have access to the raw data and are responsible for the accuracy of the data analyses.

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