

The prevalence and clinical effect of immunogenicity of TNF- α blockers in patients with axial spondyloarthritis

G. Bornstein^{1,2}, M. Lidar³, P. Langevitz³, A. Fardman², I. Ben-Zvi⁴, C. Grossman⁴

¹The Talpiot Medical Leadership Program, ²Department of Internal Medicine D, ³Rheumatology Unit, ⁴Department of Internal Medicine F, The Chaim Sheba Medical Center, Tel-Hashomer, affiliated to the Sackler Faculty of Medicine, Tel-Aviv University, Israel.

Abstract

Objective

To evaluate the prevalence of immunogenicity of TNF- α blockers in axial spondyloarthritis (SpA) patients and to assess the effect of immunogenicity on drug levels and clinical response.

Methods

Patients with axial SpA treated with either infliximab (INF), adalimumab (ADA) or etanercept (ETN) were recruited to our observational cross-sectional study. Demographic and clinical data were collected and disease activity scores were assessed. Drug trough levels and anti-drug antibodies were measured in serum samples and collected before the next administration.

Results

Thirty-nine patients with axial SpA with a mean age of 46.3 ± 12.7 (10 women) were recruited to the study (14 receiving INF, 16 ADA and 9 ETN). Patients' mean therapy duration was 50.6 months (± 46.4) and 6 (15%) of them were using MTX concomitantly with the TNF- α blockers. Anti-drug antibodies were found in 6 (15%) patients (4 with INF and 2 with ADA), all of which had undetectable drug level. No anti-drug antibodies were detected in patients treated with ETN. Immunogenicity was associated with higher BASDAI (Bath Ankylosing Spondylitis Disease Index), ASDAS-CRP (Ankylosing Spondylitis Disease Activity Score) and ASDAS-ESR.

Conclusion

Axial SpA patients failure to respond to TNF- α blockers may be at least partially related to immunogenicity. Measurement of anti-drug antibodies and drug levels in these patients may assist in determining further treatment strategies.

Key words

immunogenicity, TNF- α blockers, axial spondyloarthritis

Gil Bornstein, MD, PhD
 Merav Lidar, MD
 Pnina Langevitz, MD
 Alexander Fardman, MD
 Ilan Ben-Zvi, MD
 Chagai Grossman, MD

Please address correspondence to:
 Dr Gil Bornstein,
 Internal Medicine D,
 The Chaim Sheba Medical Center,
 Tel Hashomer 52621, Israel.
 E-mail: gilbor16@gmail.com

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Introduction

Ankylosing spondylitis (AS) and axial spondyloarthritis (Axial SpA) are two related diseases which respond well to treatment with tumour necrosis factor (TNF)- α blockers such as infliximab (INF), adalimumab (ADA), etanercept (ETN), golimumab and certolizumab (1, 2). NSAIDS are considered the first-line treatment in axial SpA, yet failure to achieve remission with these drugs necessitates upgrading treatment to biologic preparations unless local steroid injection to the sacroiliac joints is preferred. The use of synthetic disease-modifying anti-rheumatic drugs (DMARDs) such as methotrexate (MTX) and salazopyrin (SLZ) has not been proven to be effective in axial SpA (3). However, MTX may eliminate the development of anti-drug antibodies and thereby improve the efficacy of TNF- α blockers (4). Anti-drug antibody formation, which can also be referred to as immunogenicity, may be related to limited drug efficacy in patients with rheumatoid arthritis (RA), inflammatory bowel disease (IBD) and psoriatic arthritis (PsA) (5-8). It is estimated that approximately 40% of AS patients do not respond to TNF- α blockers (9). Anti-drug antibody formation is considered to be one of the explanations for decreased clinical response in AS patients, particularly for secondary, but also for primary non-responsiveness (10). Evidence is scarce regarding the association between immunogenicity and decreased response to TNF- α blockers in axial SpA patients. It has been shown that patients with AS develop anti-drug antibodies (11) more frequently than patients with RA, possibly due to the fact that MTX is not used routinely in these patients. However, previous studies have failed to demonstrate any beneficial effect from the addition of MTX to the standard treatment with infliximab in AS patients (12-15).

Thus, it is unclear whether immunogenicity is related to poor clinical response to TNF- α blockers in axial SpA patients. We therefore examined the prevalence of TNF- α -blockers immunogenicity in axial SpA patients and its association with drug levels and disease activity.

Patients and methods

Study design and patients

We conducted an observational cross sectional analysis of patients with axial SpA treated with INF, ADA or ETN in the rheumatology outpatient clinic at the Chaim Sheba Medical Center at Tel-Hashomer between January 2015 and June 2016.

All patients fulfilled the ASAS classification criteria for axial SpA and were >18 years. Their treatment protocol was either intravenous (IV) infusions of INF 5 mg/kg every 8 weeks, subcutaneous (SC) ADA 40 mg every other week or SC ETN 50 mg weekly.

The research protocol was approved by the local ethics committee and all patients gave their consent to participate in the study.

Demographic and clinical data

Data was collected by questioning the patients about comorbidities and chronic medications. Blood samples were collected before the administration of the next treatment for C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), anti-TNF- α drug level and anti-drug antibodies. Clinical assessment was done using the Ankylosing Spondylitis Disease Activity Score (ASDAS) and the Bath Ankylosing Spondylitis Disease Activity Index (BASDAI).

Measurement of TNF- α blocker levels and anti-drug antibodies

Patients' serum samples were collected and frozen in -20°C before quantification of drug and anti-drug levels. Drug concentrations were expressed in micrograms/millilitre ($\mu\text{g/ml}$). Commercial (Progenika Biopharma) promonitor-INF enzyme linked immunosorbent assay (capture ELISA) was used for quantitative determination of INF drug level. INF detection threshold was $0.035 \mu\text{g/ml}$ and levels above this range were considered positive. For the quantitative determination of ADA drug levels a commercial (Progenika Biopharma) promonitor-ADA sandwich ELISA was used. ADA detection threshold was $0.024 \mu\text{g/ml}$ and levels above this range were considered positive. Promonitor-ETN sandwich ELISA

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(Progenika Biopharma) was used for quantitative determination of ETN drug levels and drug threshold was 0.035 μ g/ml. Levels above this range were considered positive.

Anti-drug antibodies and their concentrations were expressed in absorbance units/milliliter (AU/ml). Commercial (Progenika Biopharma) promonitor-anti-INF, promonitor-anti-ADA and promonitor-anti-ETN bridging ELISAs were used for quantification of ADAb levels. Anti-INF antibodies, anti-ADA antibodies and anti-ETN antibody levels above the threshold of 5, 10 and 142 AU/ml, respectively were considered positive.

All assays were conducted according to manufacturer instructions.

Statistical analysis

All data were analysed using SPSS software 20 (SPSS, Inc., Chicago, Illinois, USA). We used one-way analysis of variance (ANOVA) with Tukey test post-hoc analysis, Student *t*-test and Chi square test in order to compare means of variables with normal distributions and proportions, respectively. A *p*-value of <0.05 was considered statistically significant.

Results

Patient characteristics

Thirty-nine patients (10 women) with a mean age of 46.3 \pm 12.7 (range 21–70) with axial SpA were recruited to our study. Table I summarises the clinical and demographic characteristics of our study cohort. The mean duration of TNF- α blocker treatment was 50.6 (\pm 46.4) months. 16 patients (41%) were using ADA, 14 patients (35%) were using INF and 9 patients (23%) were using ETN. Six patients (15%) were treated concomitantly with MTX. The mean BASDAI was 3.5 (which represents moderate to severe disease), mean ASDAS-CRP was 2.43 (high disease activity) and mean ASDAS-ESR was 2.57 (high disease activity). No significant statistical differences were demonstrated among treatment subgroups in all demographic and clinical parameters and particularly regarding disease activity scores and inflammatory markers.

Table I. Clinical and demographic data.

Variable	Etanercept	Adalimumab	Infliximab	All patients	<i>p</i> -value
Number of patients	9	16	14	39	--
Sex (M/F)	6/3	12/4	11/3	29/10	0.813
Age (years)	48.6 \pm 12.4	45.8 \pm 13.5	45.5 \pm 12.7	46.3 \pm 12.7	0.832
Tx duration (months)	64.7 \pm 40.2	33.4 \pm 24.2	61.2 \pm 62.7	50.6 \pm 46.4	0.153
MTX Tx, n (%)	1 (11.1%)	1 (6.2%)	4 (28.6%)	6 (15.4%)	0.221
BASDAI	3.7 \pm 2.6	3.46 \pm 2.3	3.44 \pm 3.05	3.5 \pm 2.6	0.971
ASDAS-ESR	2.57 \pm 1.1	2.52 \pm 1.07	2.62 \pm 1.4	2.57 \pm 1.18	0.972
ASDAS-CRP	2.53 \pm 1.26	2.27 \pm 1.32	2.54 \pm 1.38	2.43 \pm 1.3	0.82
CRP	6.3 \pm 6.24	11.06 \pm 30.4	8.5 \pm 8.4	9.06 \pm 20.04	0.85
ESR	15.7 \pm 8.6	18.8 \pm 12.4	22.7 \pm 19.6	19.5 \pm 14.7	0.53

Tx: treatment; MTX: methotrexate; ASDAS: Ankylosing Spondylitis Disease Activity Score; BASDAI: Bath Ankylosing Spondylitis Disease Activity Score; CRP: C-reactive protein; ESR: erythrocyte sedimentation rate.

Table II. Drug levels and ADAb prevalence.

	Etanercept	Adalimumab	Infliximab	All patients
Detected drug levels, n (%)	9 (100)	14 (87)	10 (71)	33 (85)
ADAb, n (%)	0 (0)	2 (13)	4 (29)	6 (15)

ADAb: anti-drug antibodies

Table III. Correlation between clinical and demographic data and the presence of ADAb.

Variable	Ab-negative (mean \pm SD)	Ab-positive (mean \pm SD)	<i>p</i> -value
Number of patients	33	6	
Age, years	47.3 \pm 12.9	41.3 \pm 11.6	0.3 ¹
Female Sex	7 (21%)	3 (50%)	0.13 ²
Tx duration (months)	54.5 \pm 48.24	29.2 \pm 28.1	0.2 ¹
MTX Tx	6 (18%)	0 (0)	0.25 ²
BASDAI	2.98 \pm 2.35	6.4 \pm 2.1	0.002¹
ASDAS-ESR	2.32 \pm 1.05	3.92 \pm 0.92	0.001¹
ASDAS-CRP	2.2 \pm 1.2	3.7 \pm 1.04	0.007¹
CRP	8.8 \pm 21.3	10.1 \pm 11.4	0.8 ¹
ESR	17.06 \pm 10.2	33 \pm 26.7	0.2 ¹

¹Student *t*-test; ²Chi square test; MTX: methotrexate; ADAb: anti-drug antibodies; Ab: antibody; ASDAS: Ankylosing Spondylitis Disease Activity Score; BASDAI: Bath Ankylosing Spondylitis Disease Activity Index; CRP: C-reactive protein; ESR: erythrocyte sedimentation rate.

Prevalence of anti-drug antibodies and drug levels

Table II presents the distribution of anti-drug antibodies and drug levels in our cohort. Six patients (15%) (4 with INF and 2 with ADA) had anti-drug antibodies. No immunogenicity was observed in ETN users. A mirror picture was notable regarding the drug levels. In all patients with anti-drug antibody drug levels were undetectable, whereas in those without anti-drug antibodies sufficient drug levels were detected. Patients who were treated with MTX did not develop anti-drug antibodies.

Association between clinical and demographic data and immunogenicity

Table III summarises the association between clinical and demographic variables and the presence of anti-drug antibodies. Significantly higher activity scores were observed among patients with anti-drug antibodies (e.g. BASDAI, ASDAS-CRP and ASDAS-ESR). Figure 1 highlights the fact that each one of disease activity scores was about 2-fold higher in patients who developed anti-drug antibodies compared with those that did not develop anti-drug antibodies. No correlation was observed between

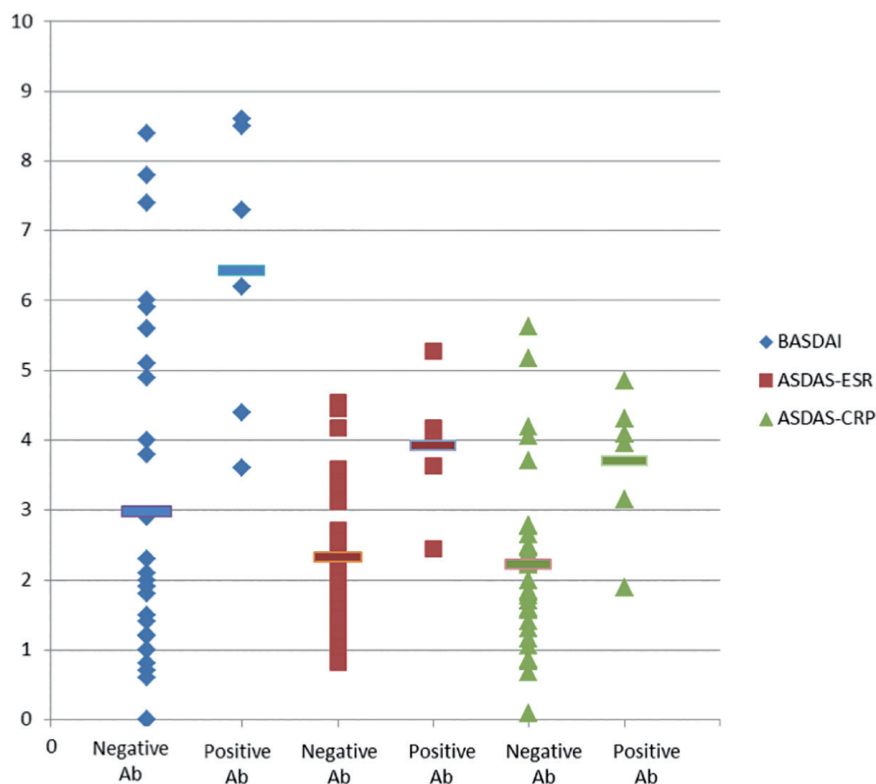


Fig. 1. Average activity scores of patients with ADAb compared to patients without ADAb. Ab: antibody; ASDAS: Ankylosing Spondylitis Disease Activity Score; BASDAI: Bath Ankylosing Spondylitis Disease Activity Index.

immunogenicity and other clinical variables in our cohort including the use of MTX, yet it can be seen that all patients who did not receive MTX did not develop anti-drug antibodies.

Discussion

TNF- α blockers are the cornerstone biological DMARD treatment for axial SpA patients with a well-established effect on disease severity and comorbidities. Primary and secondary failure of these agents to reduce disease activity constitutes a significant barrier to the treatment of patients with axial SpA since alternative effective treatments, e.g. interleukin-17 (IL-17) antibodies are limited.

In our study we found that about 20% of the patients treated with either INF or ADA developed anti-drug antibodies, whereas none of the patients treated with ETN had anti-drug antibodies. The immunogenicity was associated with undetected plasma drug levels and higher disease activity. Our findings suggest that immunogenicity may play a role in treatment failure of TNF- α

blockers among axial SpA patients. This process may also explain why some patients have a favourable initial response that fades overtime, namely secondary failure.

The rate of anti-drug antibodies in our cohort resembles results from previous studies that demonstrated an immunogenicity prevalence rate of approximately 25% in AS patients (4, 7, 16, 17). Kneepkens *et al.* (4) have demonstrated prospectively that the development of anti-drug antibodies in AS patients was inversely associated with ADA drug levels and that immunogenicity correlated with clinical response to the drug. Similar results were described in other rheumatic diseases. A recent study by Zisapel *et al.* (8) showed that the development of anti-drug antibodies in psoriatic arthritis (PsA) patients was associated with low therapeutic TNF- α blockers levels and high disease activity scores. They also demonstrated that the use of MTX in PsA patients significantly decreased the development of anti-drug antibodies and suggested that MTX use should be considered in com-

bination with TNF- α blocking agents as a mean to preserve their efficacy.

Several studies of RA patients treated with TNF- α blockers have demonstrated the existence of immunogenicity. These studies have also shown that concomitant use of MTX in these patients reduced immunogenicity rates (18-20). In our study all the patients treated with MTX did not develop anti-drug antibodies. However, we found no significant statistical difference between patients with and without anti-drug antibodies in relation to MTX use, possibly due to the relatively small study population.

Another important issue that should be addressed is the fact that none of the patients treated with ETN developed anti-drug antibodies. In this regard, it is imperative to keep in mind the basic difference that exists between the mechanism of action of ETN, which is a soluble TNF receptor and between ADA and INF which are TNF antibodies. One of the secondary effects of monoclonal antibodies is the formation of anti-drug antibodies, a phenomenon that is not expected with a soluble TNF receptor.

Our study has several clinical implications regarding the management of patients with axial SpA. First, the fact that immunogenicity is probably an important clinical issue in the preservation of TNF- α blockers efficacy along with the relatively few therapeutic alternatives in axial SpA requires determining an algorithm towards TNF- α blockers poor-responders. In these patients it seems to be worthwhile to measure anti-drug antibodies and drug levels. In cases of detected drug levels and no anti-drug antibody formation it is reasonable to switch to a different TNF- α blocker, either a TNF monoclonal antibody or either a soluble TNF receptor. Switching to an IL-17 monoclonal antibody which is a drug with a different mechanism of action should also be considered in these cases (21). In cases of anti-drug antibody formation and undetectable drug levels it would be more reasonable to switch to a different TNF- α blocker. The preference of a TNF receptor rather than a different TNF monoclonal antibody may be considered in these cases.

Although cross reactivity between anti INF antibodies and anti ADA antibodies has not been well established, a previous study has shown that antibodies against INF are associated with de novo development of antibodies to ADA and therapeutic failure in INF-ADA switchers with IBD (22). Another important clinical issue is the use of MTX in axial SpA patients. As mentioned above, previous studies have found that MTX may reduce immunogenicity rate (8, 18-20). Given the fact that MTX has not been found efficient in axial SpA, its use may be considered solely for the purpose of preserving TNF- α blockers efficacy, particularly since alternative treatment options are limited. In our study we did not find a statistically significant association between MTX use and prevalence of anti-drug antibodies. However, we did observe a trend regarding the effectiveness of MTX in reducing immunogenicity. The lack of clinical significance may be related to the small number of patients treated with MTX in our study. The use of MTX to preserve TNF- α blockers efficacy in axial SpA should be considered. In addition, the optimal dose required in these cases should be established. Large randomised controlled studies should be conducted in order to establish this mode of treatment.

Our study has several limitations. First, the study population is relatively small. Second, it is an observational cross-sectional study and not a prospective one. However, despite the small number of patients we were able to show that in patients with axial SpA the development of anti-drug antibodies to TNF- α blockers is associated with drug levels and clinical response.

It is important to emphasise that immunogenicity is not the single factor determining the response of AxSpA patients to TNF- α blockers, as the rates of response to ETN are generally comparable to those of ADA and INF. However, the results of this study support the role of immunogenicity in the clinical response of TNF- α blockers.

In conclusion, we found that among patients with axial SpA, poor response to TNF- α blockers may be related to immunogenicity. Measurement of anti-drug antibodies and drug levels in these patients may assist in determining further treatment strategies. Further large prospective studies assessing the role of MTX in preserving TNF- α blockers efficacy in axial SpA should be conducted.

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