Method Comparison Study of Two Commercially Available Tests for the Measurement of Ustekinumab Levels

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Abstract

A prospective, observational cohort study was conducted in Crohn´s disease (CD) patients treated with ustekinumab (UTK). Trough UTK and anti-UTK antibody levels were measured with Promonitor®-UTK and Promonitor® Anti-UTK tests (Progenika, Spain), respectively and compared to the corresponding results using LISA TRACKER Ustekinumab DUO (LTU) (Theradiag, France) in 100 serum samples collected at baseline and during the course of UTK treatment. Qualitative and quantitative analysis comparison of the two tests was done using descriptive statistics, Pearson Correlation, Passing-Bablok regression and Bland-Altman analysis. Both assays correlated and can be used for monitoring of UTK levels, however the measurement range and the sample dilution recommended by Promonitor-UTK covers more accurately the UTK concentrations found in CD patients, whereas use of the LTU forces the user to repeat the testing or perform two sample dilutions by default because the recommended sample dilution is not appropriate for the population. In the absence of a gold standard method to unequivocally assess tests accuracy, UTK concentration results measured with different tests should be taken into account with caution and it is advised that monitoring drug concentrations in patients over time would be best done using the same test system.

Keywords: Ustekinumab; Therapeutic Drug Monitoring; Inflammatory Bowel Disease

Abbreviations

TNF: Tumor Necrosis Factor; IBD: Inflammatory Bowel Disease; UTK: Ustekinumab; TDM: Therapeutic Drug Monitoring

Introduction

Anti-TNF biological therapies like infliximab and adalimumab are very effective in treating patients with moderate to severe IBD. However, a significant portion of patients treated with anti-TNF drugs do not response to the treatment and show either primary or secondary loss of response [1]. Ustekinumab (UTK) is a fully human immunoglobulin (Ig) G1-kappa monoclonal antibody that binds to the p40 protein subunit of the interleukin (IL)-12/23. UTK has been approved for the induction and maintenance therapy of moderate-to-severe CD patients. TDM of biological drugs is defined as the measurement of the drug and anti-drug antibodies levels to improve patient management [2-4]. TDM algorithms are mostly implemented for anti-TNF biological therapies in a reactive way, after secondary loss of response is observed [3,5,6], however recent studies have provided evidence that TDM could also be applied to other biologics like ustekinumab

and vedolizumab [7-9]. Various commercial IVD tests to monitor UTK levels in serum are available in Europe [2-4]. Here, we present a method comparison study between two commercially available CE-marked tests for the quantification of UTK levels and antibodies to UTK in serum in a cohort of IBD patients.

Methods

A prospective, observational cohort study was conducted in the IBD outpatient Clinic at the University Hospital Erlangen (Germany) in Crohn´s disease (CD) patients with treated with UTK. Trough UTK and anti-UTK antibody levels were measured with Promonitor®-UTK and Promonitor® Anti-UTK tests (Progenika, Spain), respectively and compared to the corresponding results using LISA TRACKER Usteki-numab (LTU) (Theradiag, France) in 100 serum samples collected at baseline and during the course of treatment. Both tests are based on ELISA technology and have the same intended use. The measurement range of the tests with standard dilution is 0.99 - 20 µg/mL and 0.04 - 1 µg/mL for Promonitor®-UTK and LTU, respectively. The study was performed following the Package Insert instructions provided by each manufacturer. All statistical analysis performed used a non-parametric approach (JMP software 14.0).

Results

Here, we report results for 30 patients (100% CD) recruited. UTK was administered every 8 weeks. Measurement of drug levels was performed in all patients after induction and during maintenance therapy.

Using the standard sample dilution recommended in the LTU insert (1:101), the test resulted in a low number of samples (58, 58%) out of the measurement range, which obliged to perform a reflex testing with a higher sample dilution with the consequent loss of time and reagents.

Seventy-five and 23 samples were positive and negative for UTK levels, respectively, in both Promonitor-UTK and LTU. The remaining two samples were negative for Promonitor-UTK and positive for LTU, due to the difference in the limit of detection between both assays. The quantitative comparison of the two tests was done only with the results obtained for both assays (n = 75). Median UTK levels was 2.62 µg/mL (0.19 - 15.23) and 2.92 µg/mL (0.21 - 17.67) for Promonitor-UTK and LTU, respectively (Table 1).

Descriptive statistics	LISA Tracker UTK (µg/mL)	Promonitor UTK (µg/mL)
Mean	1.68	1.89
Median	2.62	2.92
Maximum	15.23	17.67
Minimum	0.19	0.21
Standard Deviation (STD)	2.69	3.00

Table 1: Descriptive statistics of 75 patients treated with UTK.

The Pearson Correlation value was 0.956 and the average difference was 13.6% between Promonitor-UTK and LTU. Passing-Bablock regression analysis showed that UTK levels results correlated but showing a systematic proportional slight difference between the two tests (Figure 1).

The variability is proportional to concentration which can be expressed as a constant CV. Whereas agreement was good below 3 µg/mL of UTK, Bland-Altman analysis showed a systematic slight difference between both measurements above 3 µg/mL (Figure 2).
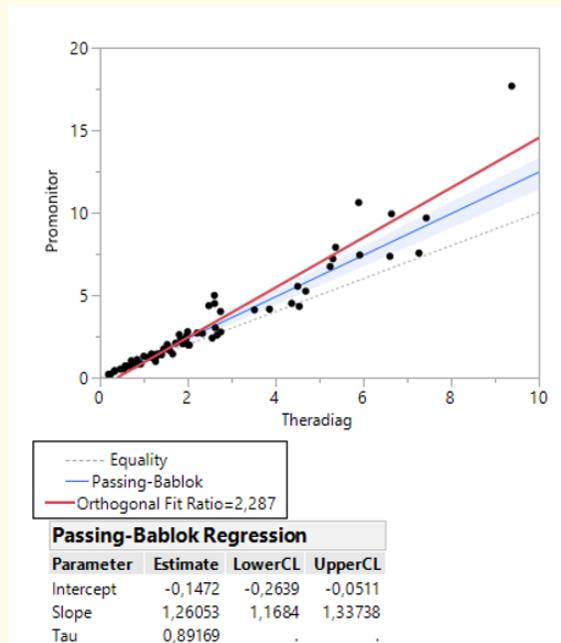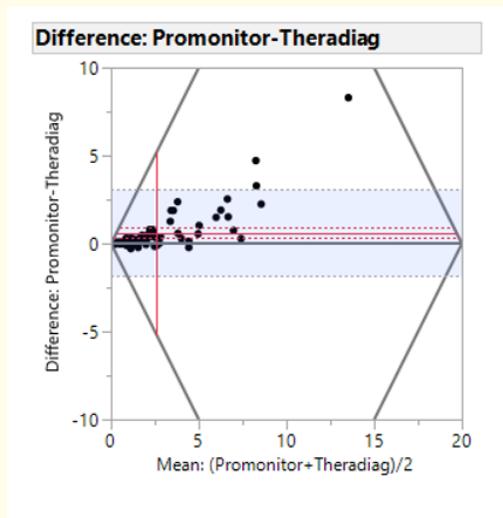
Passing-Bablok Regression

Parameter	Estimate	LowerCL	UpperCL
Intercept	-0,1472	-0,2639	-0,0511
Slope	1,26053	1,1684	1,33738
Tau	0,89169	.	.

Figure 1: Passing-Bablock of promonitor conc. (µg/mL) by Theradiag conc. (µg/mL).

Bland-Altman

Parameter	Estimate	LowerCL	UpperCL
Std Deviation	1,25008	1,04868	1,45148
Upper Limit	3,04412	2,55257	3,53567
Lower Limit	-1,8562	-2,3477	-1,3646

Figure 2: Bland-Altman of promonitor UTK conc. (µg/mL) by Theradiag conc. (µg/mL).

None of the patients in the cohort developed antibodies against UTK.

Discussion and Conclusion

UTK levels are in line with those reported in other studies in IBD [6]. There is a growing body of evidence that demonstrates the clinical utility of TDM of biologic therapy in IBD [3,5]. This is a big step towards personalized medicine and optimising the care of patients with IBD. Although there are emerging data that suggest an association between UTK drug concentrations and clinical outcomes, the sufficient evidence is still lacking to recommend TDM of UTK in clinical guidelines [3,8].

Both assays correlate and can be used for monitoring of UTK levels, however the measurement range and the sample dilution recommended by Promonitor-UTK covers more accurately the UTK concentrations found in CD patients (at week 8, median ustekinumab concentrations were 2.1 µg/mL and 6.4 µg/mL for the 130 mg and ~6 mg/kg) [7], whereas use of the LTU forces the user to repeat the testing or perform two sample dilutions by default because the recommended sample dilution is not appropriate for the IBD population.

In the absence of a gold standard method to unequivocally assess tests accuracy, UTK concentration results measured with different tests should be taken into account with caution, as a systematic slight difference between both measurements above 3 µg/mL was detected in the Bland-Altman analysis and it is advised that monitoring drug concentrations in patients over time would be best done using the same test.

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