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Antibodies-to-infliximab accelerate clearance while dose intensification reverses immunogenicity and recaptures clinical response in paediatric Crohn's disease

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Summary

Background: Antibodies to infliximab (ATI) are associated with secondary loss of response and increased risk for drug reactions. Limited studies have associated ATI with increased infliximab clearance.

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Aims: We assessed the impact of ATI on infliximab clearance and loss of response in an inception paediatric Crohn's disease cohort with 1-year follow-up.

Methods: This multi-centre prospective cohort study collected peak and trough serum infliximab/ATI concentrations from 660 infusions (78 patients) during the first year of therapy. Clinicians were blinded to these research labs. The primary outcome was the difference in infliximab clearance between ATI-positive (ATI) and ATI-negative (no-ATI) patients. Secondary outcomes included pre-treatment predictors of ATI (including HLA-DQA1 genotyping). Clinical remission, loss of response and infliximab clearance were compared between pre-ATI, during ATI and following ATI resolution with MANOVA. Time to ATI was calculated by Cox proportional Hazards model.

Results: ATI were detected in 68% (53/78) patients with a median concentration of 76 ng/mL (range 23–1828). Maximum ATI concentration was <200 ng/mL in 73.6% (39/53). Median clearance in ATI patients was higher (with higher clearance if loss of response), compared to no-ATI patients ($P < 0.001$). Neutrophil CD64 ratio >6 and starting dose <7.5 mg/kg independently predicted ATI in multivariable regression, while HLA-DQA1*05 presence did not. Dose adjustment resolved ATI in 37.5% (12/32) patients with concomitant infliximab concentration and clearance recovery. A maximum ATI level of ≤99 ng/mL predicted ATI resolution (area under the receiver operating curve 0.80 [95% CI 0.64–0.96]).

Conclusions: In this real-world cohort, ATI as low as 23 ng/mL impacted drug clearance. Our data suggest that dose optimisation for low-level ATI can improve infliximab clearance and prevent loss of response.

1 | INTRODUCTION

Although TNF-alpha antagonists (anti-TNF) have become one of the most effective medical treatments for inflammatory bowel diseases (IBDs),[1] low drug exposure can lead to anti-drug antibody formation (immunogenicity) and increase the risk of infusion reactions or loss of response.[2] As standard (label-guided) dosing with infliximab (IFX) monotherapy is associated with high rates of immunogenicity,[2] clinicians are left to decide between proactive therapeutic drug monitoring with IFX monotherapy, combination therapy (IFX and an immunomodulator), stratification of risk factors for immunogenicity with pharmacogenomics such as human leucocyte antigen (HLA) polymorphisms or a combination of these approaches to optimise efficacy and durability.[3–5]

Prior data demonstrated that episodic IFX infusions were a significant risk for anti-TNF immunisation as well as infusion reactions.[6] Whether guided by proactive or reactive therapeutic drug monitoring, IFX dose optimisations during maintenance to achieve a targeted drug concentration has been associated with mucosal healing, improved drug durability and a reduction in hospitalisations.[7–9] In a post-hoc analysis of the SONIC trial, the superior rates of mucosal healing with combination IFX therapy were likely secondary to achieving higher anti-TNF concentrations.[3] Moreover, ECCO-ESPGHAN recently published guidelines supporting the use of early proactive therapeutic drug monitoring to guide anti-TNF dosing strategies and optimise drug exposure to minimise the risk to develop anti-drug antibodies.[10]

Recent pharmacogenetic data identified that an HLA polymorphism was associated with a higher risk of immunogenicity for which commercial assays are available.[5] As an alternative to initial proactive therapeutic drug monitoring based on trough levels, pharmacogenetic testing prior to starting of IFX to stratify immunogenicity risk is now available. However, as no formal pharmacokinetic (PK) or prospective interventional studies based on HLA genotype have been completed, it is unclear how this approach fits into specific paediatric populations or therapeutic drug-monitoring guidelines.

Several population PK models and other studies have shown that IFX clearance is affected by IBD severity (extensive colitis leading to high faecal loss or TNF burden),[11] biomarkers of inflammation (serum albumin, erythrocyte sedimentation rate [ESR] or peripheral blood neutrophil Fcγ Receptor I activity ratio [nCD64])[12–14] and combination therapy (thiopurines or methotrexate).[12,13,15–17] Given the dynamic nature of these factors, there is renewed interest to more accurately predicting patient-specific IFX exposure with the use of model-informed precision dosing.[13] We and others have shown that clearance can be estimated with Bayesian PK models and more optimal therapeutic targets can be achieved with model-informed precision dosing.[13,18,19] The first IFX PK models were developed from the pivotal clinical trials and identified immunogenicity as a covariate for clearance amongst a subgroup of adult patients.[20] In a more recent real-world study, a population-based PK model was developed for children and young adults receiving IFX to predict inter-individual drug clearance with the discovery of novel covariates of clearance including antibodies to infliximab (ATI).[13]

In a recent retrospective study that included children and young adults receiving IFX,[12] we found immunogenicity was associated with clearance with a positive correlation between ATI level (binary) and increased IFX drug clearance.[12] Subsequently in our prospective, real-world paediatric Crohn's disease cohort, we found ATI (continuous) correlated with drug clearance.[13] This was also confirmed in a retrospective adult IBD paper and a biosimilar clinical trial amongst non-IBD patients.[21,22] While these models confirm that ATI is an important covariate, the impact of low and high ATI on IFX drug clearance overtime after initial development is unclear, and it is unknown how this relates to loss of response in paediatric Crohn's disease patients. Our primary objectives were to assess the impact of ATI on IFX clearance and loss of response during the first year of therapy. Secondary objectives included evaluating additional pre-treatment (clinical, biochemical and pharmacogenetic) predictors of ATI.

2 | MATERIALS AND METHODS

2.1 | Study design

This research was a post-hoc analysis of the REFINE study.[13] REFINE was a multi-centre cohort study to evaluate the PK of IFX in children and young adults with Crohn's disease.[13] While the majority of patients received label-guided (5 mg/kg) IFX induction, dose optimisations throughout the year (including induction) were at the discretion of the primary clinician. Blood samples were collected immediately prior (trough) and up to 1-hour after an infusion (peak). Stool samples were collected longitudinally, at five infusion intervals within the first therapy year to assess faecal calprotectin. Clinicians were blinded

to all research labs but could obtain drug levels independent from the study to inform their clinical decisions. Disease activity assessments were conducted at each infusion up to week 52. The study was conducted following Institutional Review Board approval at all participating medical centres including Cincinnati Children's Hospital Medical Center, Connecticut Children's Medical Center, Medical College of Wisconsin and Nationwide Children's Hospital.

2.2 | Participants

The analysis included data from 78 anti-TNF naïve participants who started IFX between August 2014 and October 2019. Eligible participant criteria included a confirmed diagnosis of Crohn's disease with established criteria[23] and <22 years of age. Patients could receive additional IBD medications such as corticosteroids and immunomodulators up to the discretion of their clinical provider. Participants with an enteric infection 2 weeks prior to IFX induction or who had prior exposure to any biological therapy were excluded. All parents/guardians and participants provided written consent and/or assent before enrolment.

2.3 | Measures and Outcomes

Age, sex, race, ethnicity, past medical history, prior medication use, Crohn's disease severity (Paris classification)[24] and routine laboratory tests were collected at baseline. Clinical disease activity with the Pediatric Crohn's Disease Activity Index (PCDAI), as well as other validated versions including the weighted PCDAI (wPCDAI),[25] and venous blood sampling for PK analysis was obtained during induction and each subsequent IFX maintenance infusion. Clinical remission was defined by a wPCDAI score <12.5 and off prednisone. Loss of response was defined as wPCDAI ≥12.5 for two consecutive infusions or starting prednisone after initial clinical remission was achieved. Biochemical remission was defined as a faecal calprotectin <250 μg/g.[26] Additionally, a more strict faecal calprotectin remission was defined as <150 μg/g.

The primary outcome was the difference in IFX clearance between patients who had incident ATI compared to clearance amongst patients who were ATI naïve within the first year of IFX treatment. In the analysis, patients with ATI within the first year were labelled as 'ATI' vs patients who did not develop ATI within the first year as 'no-ATI' regardless of when they developed ATI in that year.

Secondary outcomes included pre-treatment predictors of ATI (including HLA-DQA1 genotyping), the association of ATI with loss of response and the natural history of ATI overtime within the first IFX treatment year. Additional exploratory outcomes included ATI resolution or improvement (by intervention) overtime and the cut-off for which ATI was able to be resolved with intervention.

2.4 | ATI and IFX analyses

ATI and IFX drug concentrations were analysed from serum with the electrochemiluminescence immunoassays (ECLIA) by LabCorp/Esoterix (Calabasas Hills, CA).[27] This drug-tolerant ATI assay (up to 100 μg/mL of IFX) has a lower detection limit of 22 ng/mL. ATI titres are designated as low (22–200 ng/mL), intermediate (201–1000 ng/mL)

or high (>1000 ng/mL) by LabCorp. ATI over 200 ng/mL may be considered clinically relevant based on the high specificity for loss of response at this cutpoint.[13] Moreover, our team previously found that levels above 329 ng/mL were less likely to reverse to levels under 200 ng/mL in response to IFX dose intensification without the addition of an immunomodulator.[28]

2.5 | Neutrophil CD64 analysis

Peripheral blood was collected immediately prior to an IFX infusion and processed within 48 hours for the measurement of the neutrophil surface expression of Fcγ receptor I (CD64). The lymphocyte, monocyte and granulocyte populations are defined by their forward and side scatter characteristics with CD163 staining to further define the monocyte population and CD45, a pan-leucocyte marker. The nCD64 is the calculated ratio of the mean fluorescence intensity of granulocyte CD64 expression to the lymphocyte CD64 mean fluorescence intensity by quantitative flow cytometry (FACSCantos; BD Biosciences, San Jose, CA).[13]

2.6 | Pharmacokinetics analysis

Exposure (area under the concentration-time curve, AUC, μg h/mL) and IFX drug clearance (L/h) in individual patients were estimated using Bayesian estimation with non-linear mixed-effects modelling (NONMEM) software (Version 7.2.0, ICON Development Solutions, Ellicott City, MD). A novel, real-world paediatric PK model developed by our group was used as the Bayesian prior.[13] The PK model development and evaluation have been described and published elsewhere.[13] All PK model parameters (clearance, central and peripheral volumes of distribution, and intercompartmental clearance) were standardised by body weight as previously described.[13] The additional covariates on clearance in the PK model included the following continuous variables, serum albumin, ATI, ESR, and nCD64. Data of IFX infusion, IFX drug concentration measurements and covariate data were used for the Bayesian estimation. The AUC and clearance were estimated at the time of each concentration measurement.

2.7 | Pharmacogenetic analysis

Targeted pharmacogenetic testing was conducted to evaluate whether ATI patients were more likely to have the HLA-DQA1*05 polymorphism (1 or both alleles), as previously seen in other studies.[5,29,30] HLA-DQA1 genotyping was tested by polymerase chain reaction/sequence-specific oligonucleotide probes (DNA Identity, LabCorp, Burlington, NC). DNA was available from 51 of 78 subjects who had rich PK sampling.

2.8 | Statistical analysis

To evaluate our primary outcome, we explored the main independent variable (ATI) as both a continuous and a dichotomous measure (ATI presence vs ATI absence) to assess its influence on the primary dependent continuous variable (clearance). For continuous measures, we report means with standard deviation (SD) or medians with the 25%–75% interquartile range (IQR) based on the data distribution. The association between ATI (continuous measure) and clearance was assessed using the Spearman correlation with a

scatterplot included. Non-parametric statistics for single comparisons (Wilcoxon rank sum) and Kruskal-Wallis test with post-hoc Dunn's were used for multiple comparisons. The Wilcoxon rank sum test was used for ATI and loss of response as a dichotomous scale for our secondary outcomes. Univariable and subsequent multivariable logistic regression were performed to evaluate predictors of ATI vs non-ATI development. The multivariable model was selected with the Bayesian Information Criterion (BIC) method ('leaps' package using *R* version 4.0.2, The R foundation for Statistical Computing, Vienna, Austria). A final model was chosen utilising a BIC plot and selection table, which included two significant variables 'starting dose <7.5 mg/kg' and 'nCD64 >6'. Hazard ratios (HRs) for the time to develop ATI were estimated by Cox regression analysis with the 'survival' and 'survminer' packages. The Dunn's test calculated the effect of ATI overtime (pre-ATI, when ATI were present and when ATI were resolved) on clearance. Multivariate analysis of variance (MANOVA) calculated the effect of ATI overtime (at the infusion prior to ATI development, at the infusions when ATI were present and at the infusion when ATI resolved) on clearance when stratified by the loss of response status which was depicted with mean lines of spaghetti plots ('ggplot2' package). Finally, receiver operator characteristics (ROC) curve analysis and the Youden J statistic were conducted with the 'OptimalCutpoints' package within R to determine the ATI cutpoint associated with successful resolution of ATI.

3 | RESULTS

The cohort included 78 patients who received a total of 660 IFX infusions (mean 8.7, SD 3.7), over the 1-year observation. ATI were detected in 53/78 (68%) patients during the first year of IFX treatment. The majority of patients developed ATI in the maintenance phase with six ATI events detected at infusion 4 and only three detected during induction (all three at infusion 3). Amongst these 53 ATI events, the ATI concentration range was 23–1828 ng/mL with 14 (26.4%) ATI levels >200 ng/mL. Baseline demographics, disease severity and concomitant medication use were not statistically significantly different between patients who developed ATI compared to the patients who did not (no-ATI, Table 1). Specifically, immunomodulator use was low in both groups (2/53 and 3/25 patients, in the ATI and no-ATI groups, respectively). However, while not statistically significantly different, ATI patients were numerically younger than no-ATI patients (median age of 11.5 years [IQR 9–15] compared to 14 years [IQR 11–15, $P = 0.08$]). Overall, throughout the first year, ATI patients had a higher median clearance of 0.0111 L/h than no-ATI patients of 0.0094 L/h ($P < 0.001$).

3.1 | Baseline laboratory testing, drug exposure and ATI

We found the baseline nCD64 activity ratio was higher amongst ATI patients (median 6.6, IQR 6.2–7.2, n = 50) vs no-ATI patients (median 6.2, IQR 4.7–6.9, n = 23, $P = 0.04$). While baseline median faecal calprotectin was not significantly different between ATI (1599 μg/g, IQR 1126–2501, n = 34) and no-ATI patients (2330 μg/g, IQR 1205–2501, $n = 18$, $P = 0.39$). Faecal calprotectin at the end of induction trended higher in ATI patients (708.5 μg/g, IQR 373.8–2473.8) compared to no-ATI patients (349 μg/g, IQR 75.3–1133.8, $P = 0.06$). At the end of induction, 21% of ATI patients were in calprotectin remission (<250 μg/g) compared to 44% of no-ATI patients ($P = 0.14$). When a stricter calprotectin definition was used (<150

μg/g), 9% of ATI patients were in strict calprotectin remission compared to 33% of no-ATI patients ($P = 0.05$).

As ATI risk is known to increase with low drug exposure, it was not surprising to find that ATI patients had less cumulative IFX induction exposure, as measured by AUC week 0–14 of 69 308.5 μg h/mL (IQR 54 431–101 140) vs no-ATI patients (102 820 μg h/mL, IQR 67 693–125 420, $P = 0.005$). End of induction IFX trough levels was also lower amongst ATI patients 4.9 μg/mL (IQR 2.4–9.7) vs no-ATI patients 9.6 μg/mL (IQR 3.6–15, $P = 0.04$).

3.2 | Immunogenicity and IFX Clearance

When clearance was explored amongst all infusions, even patients with the lowest detected ATI level (23 ng/mL) at an infusion had a significantly higher clearance than patients without ATI. Amongst infusion visits in which patients had detectable ATI, we found clearance continued to increase with higher ATI levels ($\rho = 0.37$, $P < 0.001$, Figure 1).

3.3 | Predictors of ATI development

Predictors of ATI development were first evaluated using a univariable analysis (Table 2) for baseline variables including patient- and disease-related factors, concomitant medication exposure, age of diagnosis and disease duration. In a multivariable model, *n*CD64 ratio >6 at baseline (odds ratio [OR] 6.23, 95% CI 1.80–23.54, $P = 0.005$), and IFX starting dose <7.5 mg/kg (OR 5.47, 95% CI 1.53–21.02, $P = 0.01$) were both independent predictors for ATI development in the first year of IFX therapy (Table 2). Of note, there was no difference in initial clearance between patients who had a starting dose <7.5 vs ≥7.5 mg/kg.

3.4 | Time to ATI development

The risk of ATI development over time (in days), was evaluated for each of the risk factors that were significant on the univariable regression analysis (Table 3 and Figure 2). An nCD64 >6 had an HR of 2.55 (95% CI 1.14–5.70, $P = 0.02$). Moreover, adequate IFX exposure, expressed as a week 0–14 AUC 79 348 μg h/mL,[13] was protective of ATI development over time (HR 0.48, 95% CI 1.14–5.70, $P = 0.02$).

3.5 | Pharmacogenetics and ATI

HLA-DQA1 status, in particular, the HLA-DQA1*05 polymorphism was assessed between the ATI and no-ATI groups. Amongst the subset of patients with DNA available, 45% (15/33) of patients that developed ATI had the *05 variant vs 56% (10/18) of patients who did not develop ATI ($P = 0.69$). Similarly, there was no difference in the frequency of the *05 variants between ATI groups when the ATI cut-off was set at >200 ng/mL. When we assessed the ATI group by maximum ATI concentration in this subset with HLA-DQA1 genotyping, most patients had ATI 22–200 ng/mL (82%, 27/33). There were an additional three patients (9%) with ATI between 200–500 ng/mL and three patients (9%) >500 ng/mL. For patients within the 22–200 ng/mL level, 41% (11/27) had the *05 variant, while 67% (4/6) of those >200 ng/mL had the*05 variant ($P = 0.48$). Time to ATI development was not different when stratified by HLA-DQA1*05 status in a Cox regression analysis. Immunomodulators were started in one patient with the *05 variant and one patient without the variant. Furthermore, there was no difference between faecal calprotectin, loss

of response or PK parameters (including IFX clearance and AUC) when patients with and without the *05 variants were evaluated (data not shown). In addition, there were no significant differences in immunogenicity when we evaluated HLA-DQA1*01, 02, 03 or 04 presence.

3.6 | Longitudinal trends of ATI

In this real-world cohort, 79.5% received labelled (<7.5 mg/kg at 0, 2 and 6 weeks) IFX dosing and 94% received monotherapy during induction. To describe the longitudinal history of ATI and their relations to clinical and PK outcomes, PK test results were subsequently classified into three phases: infusion prior to ATI development, infusion when ATI were present and the infusion after which ATI resolved. We found the median IFX clearance (0.0122 L/h, IQR 0.0098–0.0170) at the infusion with ATI was significantly higher compared to the median IFX clearance (0.0103 L/h, IQR 0.0083–0.0123) at the infusion prior to ATI development ($P < 0.001$). More strikingly, amongst the patients who had subsequent ATI resolution, the median clearance (0.0099 L/h, IQR 0.0084–0.0140) was also lower ($P = 0.007$) compared to the clearance at the initial ATI discovery.

Expectantly, we found that there was a similar trend between IFX concentrations and ATI phases. The median pre-ATI IFX level was 4.9 μg/mL (IQR 2.5–9.7) prior to ATI detection and 3.1 μg/mL (IQR 1.5–7.0, $P = 0.005$) while ATI were present. With resolution of ATI, the median IFX increased to 8.9 μg/mL (IQR 7.7–14.0, $P < 0.001$, Figure 3A).

Overall, there was no difference in clinical loss of response between ATI patients (42%; 22/53) vs no-ATI patients (60%, 15/25, $P = 0.20$). However, the median ATI 113 ng/mL (IQR 63–209) were higher amongst patients with loss of response compared to those without loss of response with a median ATI of 69 ng/mL (IQR 39–109, $P < 0.001$). Additionally, patients with loss of response also had a higher median clearance (0.0157 L/h, IQR 0.012–0.019) compared to those without loss of response (0.0114 L/h, IQR 0.009–0.014, $P < 0.001$). Subsequently, clearance overtime stratified by the loss of response clinical status demonstrated that patients with loss of response had a higher clearance than patients without loss of response prior to and during ATI (Figure 3B).

As this was an observational study, IFX regimen optimisations were not standardised. Amongst the cohort that had repeat ATI testing available, 68% (32/47) had undergone IFX optimisation in response to ATI (blinded to the research drawn IFX concentrations). IFX optimisation interventions included IFX interval shortening (n = 9), dose escalation (n = 13), both (n = 10) and/or addition of combination therapy with methotrexate (n = 3). ATI completely resolved (<22 ng/mL) in 37.5 % (12/32) who had IFX dose adjustment after ATI development, while there was no resolution of (0/15) ATI in patients without an IFX dose adjustment ($P = 0.005$). Of note, the median peak ATI (maximum level) amongst patients with IFX optimisation was 112 ng/mL (IQR 23–1803) vs a median of 103 ng/mL (IQR 23–1309) amongst patients without IFX optimisation. All 12 patients with complete ATI resolution received either a dose increase and/or interval shortening. From initial detection, full resolution of ATI was complete by a median of four infusions (IQR 1.75–5.25). Interestingly, none of the three patients who started concomitant methotrexate in addition to IFX optimisation had a full resolution of their ATI (Table 4).

To evaluate the likelihood of ATI resolution amongst patients with IFX optimisation, ROC analyses were conducted for the highest detected ATI level for each patient. A maximum ATI level of 99 ng/mL predicted immunogenicity resolution (<22 ng/mL), with an optimised sensitivity of 75% and specificity of 70% (area under the ROC curve 0.80, 95% CI 0.64–0.95, Figure 4). In this analysis, the sensitivity reached 100% for immunogenicity resolution at a cut-off of 197 ng/mL, however, this is at the cost of a reduced specificity (45%).

4 | DISCUSSION

Using data from a cohort of paediatric Crohn's disease patients largely receiving IFX monotherapy, we demonstrated prospectively that IFX immunogenicity is common and is associated with higher drug clearance, even at relatively low ATI concentrations (<200 ng/mL). We found independent associations between ATI development and an elevated baseline nCD64 >6, a starting dose <7.5 mg/kg and low IFX exposure (AUC 79 348 μg h/mL) during induction. Interestingly, we did not find that the HLA-DQA1*05 polymorphism was associated with ATI in our cohort. We also describe the natural history of ATI development overtime within the first year of IFX treatment with predominant monotherapy and its association with secondary loss of response. Moreover, this study identified that drug clearance increases when ATI develop and found that both clinical response and drug clearance can be recaptured with a resolution of ATI, even if ATI led to the initial loss of response.

While it is well established that ATI contributes to secondary loss of response,[2,6] our group and others previously suggested that this may be related to increased drug clearance due to immunogenicity.[12,13,21] Our current study describes the natural history of ATI development in a cohort that received predominantly labelled IFX monotherapy. A prior adult study reported on the temporal evolution of ATI within the first year of IFX therapy in relation to disease outcomes yet did not report IFX clearance.[31] Establishing these temporal relationships with clearance measures are critical to achieve optimal exposure response and to be able to intervene with early dose adjustments, such as guided by model-informed precision dosing clinical decision support tools for patients at risk for loss of response.

The post-hoc analysis from the SONIC trial has now confirmed that improved outcomes with combination immunomodulators were the result of improved PK including higher drug concentrations and lower ATI.[3] Furthermore, the adult patient population that comprised a portion of the initial IFX PK analysis only identified the presence or absence to be associated with clearance rather than the intensity of the ATI concentration.[20] Notably, our current study utilised a highly sensitive drug-tolerant ATI assay that detects lower states of immunogenicity (both bound and unbound to IFX). While prior studies assumed that low-level ATI in the presence of adequate IFX levels may be transient circulating non-neutralising ATI,[32] our current study suggests that even lower levels of ATI are related to higher clearance and loss of response. Moreover, we found that only 3 out of 14 patients with levels >200 ng/mL had decreased their ATI level below that cut-off. On the other hand, patients whose maximum ATI level throughout the year was <99 ng/mL were more likely to resolve ATI if subsequent IFX dose adjustment occurred. These results suggest that for

the sensitive assay used in this study, even lower ATI levels may need to be addressed to decrease the clearance and prevent loss of response.

Unlike the low immunogenicity rate observed in the REACH trial (3%), our real-world paediatric study, with an immunogenicity rate of 68%, yielded similar results compared to the recent large British real-world PANTS study.[33] The PANTS study, which also included paediatric patients, overall found an immunogenicity rate of 63% when tested by another drug-tolerant ATI assay. While PANTS was a very large observational cohort study in which they found that IFX immunogenicity was associated with an HLA gene polymorphism (HLA-DQA1*05),[5] they only speculated that immunogenicity formation was related to high drug clearance but did not report clearance based on sparse PK sampling.[33]

Prior analyses from the PANTS cohort and a Canadian cohort found that the HLA-DQA1*05 polymorphism was associated with anti-TNF immunogenicity with a hazard ratio between 1.9 and 7.29 amongst IBD patients.[5,29,30] In our post-hoc analysis, we did not find HLA polymorphism differences between ATI and no-ATI patients. While our study sample size was smaller than the PANTS cohort, our rich PK sampling throughout the year and drug-tolerant ATI assay could have detected (low level) ATIs in more patients, however, the ATI incidence rates were similar. While there was no difference in ATI development and HLA-DQA1*05, we did find 4/6 patients with ATI >200 ng/mL had the HLA-DQA1*05 polymorphism and it is plausible that a much larger and racially diverse cohort is needed to detect significant differences in these polymorphisms. Alternatively, larger studies are needed to determine if proactive therapeutic drug monitoring and subsequent IFX optimisation can overcome pharmacogenetic risk factors.

In our Cox regression analysis, we found time to ATI development was significantly associated with an elevated baseline nCD64 and was less likely if there was an adequate induction exposure as measured by week 0–14 AUC. This was not surprising as our group previously described that these factors were significant contributors to IFX drug clearance.[13] In contrast, other baseline biomarkers of inflammatory burden including albumin, C-reactive protein (CRP) and ESR were not different between ATI and no-ATI patients (Table 1). The primary route of IFX clearance is complex and includes intracellular proteolytic catabolism via the reticuloendothelial system, target-mediated (binding to TNFα) and binding to the Fcγ receptors such as FcγRI (CD64) which is upregulated on activated neutrophils during inflammation.[15] Furthermore, reducing mAb/Fcγ receptor binding affinity was shown to slow drug clearance.[34] While another study identified that polymorphisms in FcγRIIIa were associated with altered responses to IFX in Crohn's disease,[35] and others have shown that CD64 modulates the inhibitory activity of IFX in vitro and ex vivo, no other groups have studied nCD64 in relation to IFX clearance in Crohn's disease patients to our knowledge. Further external validation of nCD64 as a covariate for IFX clearance is warranted.

In this observational study, ATIs resolved in only 12/53 ATI-positive patients with all 12 receiving IFX optimisation. While prior studies have suggested that low-level ATIs are transient, we did not observe ATI resolution without intervention.[32] In addition, our longitudinal study design with frequent ATI sampling allowed us to appreciate temporary

ATI resolution with ATI reappearing at subsequent infusions (an important clinical observation to consider as therapeutic drug-monitoring guidelines are updated).

The results of our current study suggest that early therapeutic drug exposure and timely immunogenicity detection is important to sustain therapeutic response for these critical medications. While earlier paediatric studies have focused on improving immunogenicity after the development of ATI with dose escalation or the addition of combination therapy with immunomodulators,[28,36] our current study provides a rationale for early therapeutic drug monitoring and possibly even dose escalation based on low drug exposure or detection of increased clearance prior to ATI development. As calculating clearance without access to PK clinical decision support tools can be impractical at the bedside, routine use of blood and stool inflammatory markers and assuring adequate drug exposure during induction may be protective for ATI development. Alternatively, a practical approach could be to start combination therapy with an immunomodulator at IFX initiation. The COMBINE study (clinicaltrials.gov NCT02772965) is currently assessing the time to treatment failure (trough week 156) between combination (anti-TNF and methotrexate) and anti-TNF monotherapy in children with Crohn's disease. Until more definitive data are available, low IFX exposure during induction is a significant risk factor for immunogenicity and underscores the importance of early therapeutic drug monitoring for patients starting anti-TNF monotherapy.

Given the complexity of IFX clearance and the limited therapeutic options for children with Crohn's disease, more intensive PK and pharmacodynamic monitoring with real-time PK clinical decision support tools may be needed to identify those patients at risk for high drug clearance and provide an earlier intervention to prevent ATI or loss of response.[13] As this observational study was conducted prior to widespread use of proactive therapeutic drug monitoring across all participating centres, the majority of our cohort received standard FDA-labelled IFX monotherapy induction dosing (<7.5 mg/kg) and may have led to inadequate induction exposure to overcome long-term persistent immunogenicity.[37] While paediatric IFX monotherapy is not yet supported in the European ECCO-ESPGHAN guideline,[10] proactive therapeutic drug monitoring with initial monotherapy is a common paediatric practice in other parts of the world including North America.[4,28,38] More studies, such as the REMODEL trial (clinicaltrials.org NCT04974099), are needed to determine if dose optimisation from the start of IFX and proactive therapeutic drug monitoring can improve drug durability and endoscopic healing. We recognise that while clinical and biochemical outcomes in our study were assessed, it was not feasible to perform the gold standard endoscopic evaluation to directly assess mucosal healing. However, as a surrogate for endoscopy, we used a clinically relevant definition for loss of response and identified differences in clearance using this definition. Last, while PK studies are at risk of having limited generalizability due to quantitative differences between assays, the IFX/ATI assay used in this study has previously demonstrated strong agreement with similar assays.[39]

In conclusion, this observational prospective PK analysis demonstrated the natural history of clearance in relationship to IFX immunogenicity in a standard-of-care paediatric Crohn's disease cohort who predominantly received IFX monotherapy. More specifically, this study characterised that clearance is increased even prior to, as well as during, ATI development and is associated with loss of response. Moreover, we found that ATI

previously considered ‘low or clinically insignificant’ were associated with increased clearance and loss of response. While our cohort did not support that the HLA-DQA1 polymorphism was associated with ATI development, we did demonstrate that elevated nCD64 and low drug exposure during induction were associated with an increased risk of immunogenicity. These data underscore the need for further prospective interventional trials that examine the feasibility and benefit of proactive model-informed precision dosing and/or pharmacogenetics and novel biomarkers to individualise drug exposure as part of a treat-to-target approach to prevent secondary loss of response.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

REFERENCES

1. Rosen MJ, Dhawan A, Saeed SA. Inflammatory bowel disease in children and adolescents. JAMA Pediatr. 2015;169:1053–1060. [PubMed: 26414706]
2. Baert F, Noman M, Vermeire S, et al. Influence of immunogenicity on the long-term efficacy of infliximab in Crohn’s disease. N Engl J Med. 2003;348:601–608. [PubMed: 12584368]

3. Colombel J-F, Adedokun OJ, Gasink C, et al. Combination therapy with infliximab and azathioprine improves infliximab pharmacokinetic features and efficacy: a post hoc analysis. Clin Gastroenterol Hepatol. 2019;17:1525–1532.e1. [PubMed: 30267864]
4. Lyles JL, Mulgund AA, Bauman LE, et al. Effect of a practice-wide anti-TNF proactive therapeutic drug monitoring program on outcomes in pediatric patients with inflammatory bowel disease. Inflamm Bowel Dis. 2021;27:482–492. [PubMed: 32448898]
5. Sazonovs A, Kennedy NA, Moutsianas L, et al. HLA-DQA1*05 Carriage associated with development of anti-drug antibodies to infliximab and adalimumab in patients with Crohn's disease. Gastroenterology. 2020;158:189–199. [PubMed: 31600487]
6. Maser EA, Villela R, Silverberg MS, et al. Association of trough serum infliximab to clinical outcome after scheduled maintenance treatment for Crohn's disease. Clin Gastroenterol Hepatol. 2006;4:1248–1254. [PubMed: 16931170]
7. Ungar B, Levy I, Yavne Y, et al. Optimizing Anti-TNF-α therapy: serum levels of infliximab and adalimumab are associated with mucosal healing in patients with inflammatory bowel diseases. Clin Gastroenterol Hepatol. 2016;14:550–557.e2. [PubMed: 26538204]
8. Papamichael K, Vajravelu RK, Vaughn BP, et al. Proactive infliximab monitoring following reactive testing is associated with better clinical outcomes than reactive testing alone in patients with inflammatory bowel disease. J Crohns Colitis. 2018;12:804–810. [PubMed: 29590345]
9. Vaughn BP, Martinez-Vazquez M, Patwardhan VR, et al. Proactive therapeutic concentration monitoring of infliximab may improve outcomes for patients with inflammatory bowel disease: results from a pilot observational study. Inflamm Bowel Dis. 2014;20:1996–2003. [PubMed: 25192499]
10. van Rheenen PF, Aloi M, Assa A, et al. The medical management of paediatric Crohn's disease: an ECCO-ESPGHAN guideline update. J Crohns Colitis. 2021;15:171–194.
11. Brandse JF, van den Brink GR, Wildenberg ME, et al. Loss of infliximab into feces is associated with lack of response to therapy in patients with severe ulcerative colitis. Gastroenterology. 2015;149:350–5.e2. [PubMed: 25917786]
12. Bauman LE, Xiong YE, Mizuno T, et al. Improved population pharmacokinetic model for predicting optimized infliximab exposure in pediatric inflammatory bowel disease. Inflamm Bowel Dis. 2020;26:429–439. [PubMed: 31287855]
13. Xiong YE, Mizuno T, Colman R, et al. Real-world infliximab pharmacokinetic study informs an electronic health record-embedded dashboard to guide precision dosing in children with Crohn's disease. Clin Pharmacol Ther. 2021;109:1639–1647. [PubMed: 33354765]
14. Minar P, Jackson K, Tsai Y-T, et al. Validation of neutrophil CD64 blood biomarkers to detect mucosal inflammation in pediatric Crohn's disease. Inflamm Bowel Dis. 2017;24:198–208. [PubMed: 29272485]
15. Ordás I, Mould DR, Feagan BG, et al. Anti-TNF monoclonal antibodies in inflammatory bowel disease: pharmacokinetics-based dosing paradigms. Clin Pharmacol Ther. 2012;91:635–646. [PubMed: 22357456]
16. Rosen MJ, Minar P, Vinks AA. Review article: applying pharmacokinetics to optimise dosing of anti-TNF biologics in acute severe ulcerative colitis. Aliment Pharmacol Ther. 2015;41:1094–1103. [PubMed: 25809869]
17. Bar-Yoseph H, Pressman S, Blatt A, et al. Infliximab-tumor necrosis factor complexes elicit formation of anti-drug antibodies. Gastroenterology. 2019;157:1338–1351.e8. [PubMed: 31401142]
18. Strik AS, Löwenberg M, Mould DR, et al. Efficacy of dashboard driven dosing of infliximab in inflammatory bowel disease patients: a randomized controlled trial. Scand J Gastroenterol. 2021;56:145–154. [PubMed: 33290108]
19. Eser A, Primas C, Reinisch S, et al. Prediction of individual serum infliximab concentrations in inflammatory bowel disease by a Bayesian dashboard system. J Clin Pharmacol. 2018;58:790–802. [PubMed: 29381220]
20. Fasanmade AA, Adedokun OJ, Blank M, et al. Pharmacokinetic properties of infliximab in children and adults with Crohn's disease: a retrospective analysis of data from 2 phase III clinical trials. Clin Ther. 2011;33:946–964. [PubMed: 21741088]

21. Brandse JF, Mould D, Smeekes O, et al. A real-life population pharmacokinetic study reveals factors associated with clearance and immunogenicity of infliximab in inflammatory bowel disease. Inflamm Bowel Dis. 2017;23:650–660. [PubMed: 28195852]
22. Eser A, Reinisch W, Schreiber S, et al. Increased induction infliximab clearance predicts early antidrug antibody detection. J Clin Pharmacol. 2021;61:224–233. [PubMed: 32905628]
23. Levine A, Koletzko S, Turner D, et al. ESPGHAN revised Porto criteria for the diagnosis of inflammatory bowel disease in children and adolescents. J Pediatr Gastroenterol Nutr. 2014;58:795–806. [PubMed: 24231644]
24. Levine A, Griffiths A, Markowitz J, et al. Pediatric modification of the Montreal classification for inflammatory bowel disease: the Paris classification. Inflamm Bowel Dis. 2011;17:1314–1321. [PubMed: 21560194]
25. Turner D, Levine A, Walters TD, et al. Which PCDAI version best reflects intestinal inflammation in pediatric Crohn disease? J Pediatr Gastroenterol Nutr. 2017;64:254–260. [PubMed: 27050050]
26. Lin J-F, Chen J-M, Zuo J-H, et al. Meta-analysis: fecal calprotectin for assessment of inflammatory bowel disease activity. Inflamm Bowel Dis. 2014;20:1407–1415. [PubMed: 24983982]
27. Kelly C, Imir M, Jeanne E, et al. P058 Serum infliximab and corresponding anti-infliximab antibody: analysis of over 30,000 patient results using lab developed chemiluminescent immunoassays. Am J Gastroenterol. 2019;114:S16.
28. Colman RJ, Portocarrero-Castillo A, Chona D, et al. Favorable outcomes and anti-TNF durability after addition of an immunomodulator for anti-drug antibodies in pediatric IBD patients. Inflamm Bowel Dis. 2021;27:507–515. [PubMed: 32426829]
29. Powell Doherty RD, Liao H, Satsangi JJ, et al. Extended analysis identifies drug-specific association of 2 distinct HLA class II haplotypes for development of immunogenicity to adalimumab and infliximab. Gastroenterology. 2020;159:784–787. [PubMed: 32275970]
30. Wilson A, Peel C, Wang Q, et al. HLADQA1*05 genotype predicts anti-drug antibody formation and loss of response during infliximab therapy for inflammatory bowel disease. Aliment Pharmacol Ther. 2020;51:356–363. [PubMed: 31650614]
31. Ungar B, Chowers Y, Yavzori M, et al. The temporal evolution of antidrug antibodies in patients with inflammatory bowel disease treated with infliximab. Gut. 2014;63:1258–1264. [PubMed: 24041539]
32. Vande Casteele N, Gils A, Singh S, et al. Antibody response to infliximab and its impact on pharmacokinetics can be transient. Am J Gastroenterol. 2013;108:962–971. [PubMed: 23419382]
33. Kennedy NA, Heap GA, Green HD, et al. Predictors of anti-TNF treatment failure in anti-TNF-naive patients with active luminal Crohn's disease: a prospective, multicentre, cohort study. Lancet Gastroenterol Hepatol. 2019;4:341–353. [PubMed: 30824404]
34. Hoffmeyer F, Witte K, Schmidt RE. The high-affinity Fc gamma RI on PMN: regulation of expression and signal transduction. Immunology. 1997;92:544–552. [PubMed: 9497497]
35. Louis E, El Ghoul Z, Vermeire S, et al. Association between polymorphism in IgG Fc receptor IIIa coding gene and biological response to infliximab in Crohn's disease. Aliment Pharmacol Ther. 2004;19:511–519. [PubMed: 14987319]
36. Cohen RZ, Schoen BT, Kugathasan S, et al. Management of Anti-drug Antibodies (ADA) to biologic medications in children with inflammatory bowel disease. J Pediatr Gastroenterol Nutr. 2019;69:551–556. [PubMed: 31335833]
37. Jongsma MME, Winter DA, Huynh HQ, et al. Infliximab in young paediatric IBD patients: it is all about the dosing. Eur J Pediatr. 2020;179:1935–1944. [PubMed: 32813123]
38. Lega S, Phan BL, Rosenthal CJ, et al. Proactively optimized infliximab monotherapy is as effective as combination therapy in IBD. Inflamm Bowel Dis. 2019;25:134–141. [PubMed: 29868777]
39. Marini JC, Sendecki J, Cornillie F, et al. Comparisons of serum infliximab and antibodies-to-infliximab tests used in inflammatory bowel disease clinical trials of remicade(R). Aaps J. 2017;19:161–171. [PubMed: 27600137]

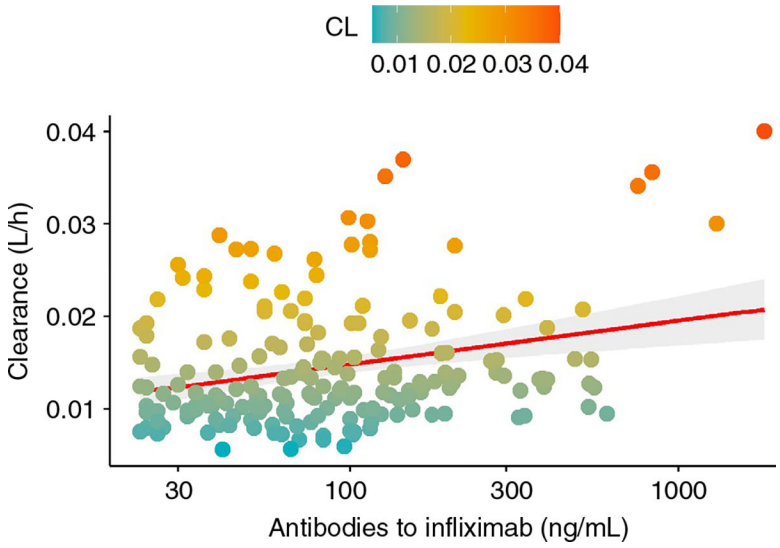

FIGURE 1.

The correlation between continuous antibodies to infliximab concentration and infliximab clearance

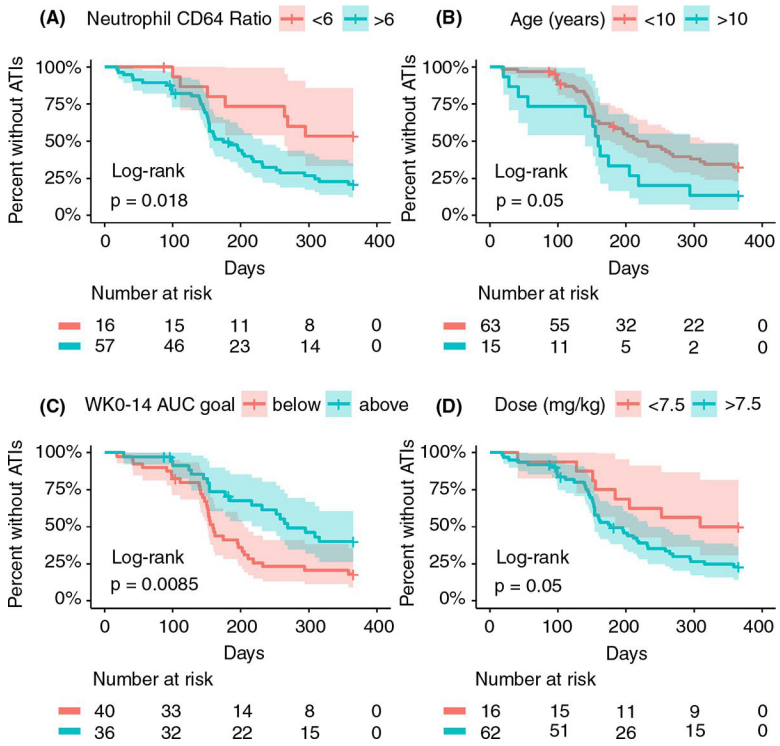

FIGURE 2.

Cox regression analyses of time to antibody to infliximab (ATI) development for (A). Neutrophil CD64 activity ratio (nCD64), (B) Patient age (years), (C) week 0–14 exposure (AUC, area under concentration curve) goal (defined by a week 0–14 AUC of 79 348 μg h/mL) and (D) the infliximab starting dose (mg/kg)

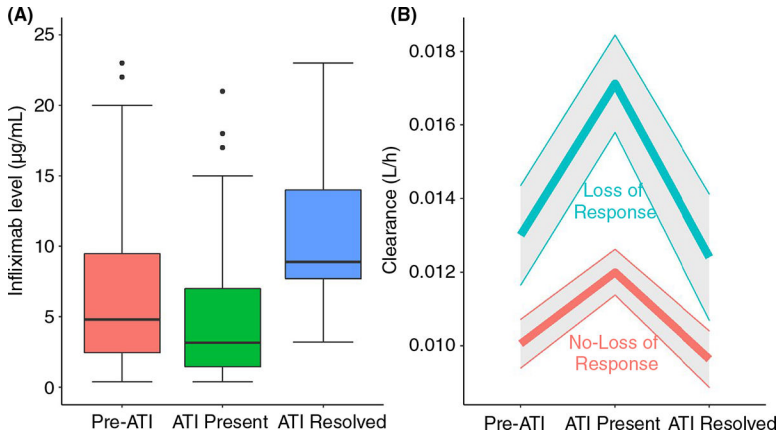

FIGURE 3.

A, The relationship between infliximab through concentration and antibodies to infliximab (ATI) evolution overtime. B, The association between infliximab clearance (L/h) and ATI overtime and further classified by loss of response status

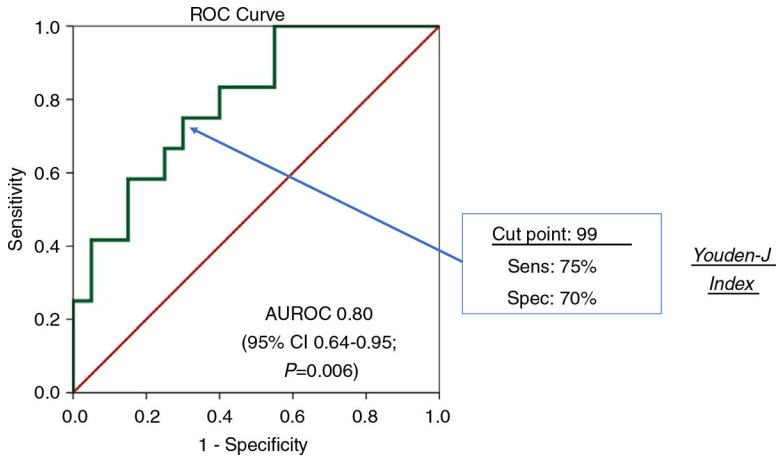

FIGURE 4.

Receiver operator characteristic (ROC) analysis of the highest antibodies to infliximab concentration that was associated with immunogenicity resolution. AUROC, area under the receiver operating characteristic curve

TABLE 1

Baseline characteristics between patients with antibodies to infliximab (ATI) and patients without ATI (no-ATI)

Baseline characteristics	ATI (n = 53)	No-ATI (n = 25)	*P* value
Age at Diagnosis, median (IQR), y	11.5 (9–15)	14 (11–15)	0.08
Sex, male, n (%)	36 (68)	14 (56)	0.44
Crohn's disease duration at induction, median (IQR), d	53 (19–422)	47 (16–214)	0.46
Paris classification (n)			
A1b (10 to <17 years)	27	13	n.s.
L1, L2, L3	4:2:42	2:3:13	
B1, B2, B3	41:4:3	14:3:1	
Steroid use at baseline, n (%)	33 (62)	17 (68)	0.81
Immunomodulator use, n (%)	2 (4)	3 (12)	0.32
ATI level, median (IQR) (ng/mL)	76 (43–141)	–	–
Baseline faecal calprotectin, median (IQR) (μg/g)	1599 (1126–2501)	2330 (1205–2501)	0.39
Baseline albumin, median (IQR) (g/dL)	3.3 (3–3.7)	3.4 (3.1–3.8)	0.35
Baseline CRP, median (IQR) (mg/dL)	1.07 (0.33–2.41)	1.00 (0.29–2.14)	0.47
Baseline ESR, median (IQR) (mm/h)	17.5 (10–28.3)	10 (8–20)	0.13
Baseline nCD64, ratio median (IQR)	6.6 (6.2–7.2)	6.2 (4.7–6.9)	0.04
Baseline wPCDAI, median (IQR)	37.5 (20–65)	35.0 (20–55)	0.95
Starting dose, median (IQR) (mg/kg)	6.0 (5.0–6.7)	6.1 (5.5–8.4)	0.09

Abbreviations: CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; IQR, interquartile range; nCD64, neutrophil CD64 activity ratio; wPCDAI, weight paediatric Crohn's disease activity index.

TABLE 2

Univariable and multivariable baseline predictors of antibodies to infliximab (ATI) development

	Univariable			Multivariable		
	OR	**95% CI**	***P* value**	**OR**	**95% CI**	***P* value**
Age <10 years	3.74	0.92–25.3	0.10	–	–	–
Starting dose <7.5 mg/kg	2.65	0.85–8.33	0.09	5.47	1.53–21.02	0.01
Baseline neutrophil CD64 activity ratio >6	3.95	1.25–13.03	0.02	6.23	1.80–23.54	0.005
Baseline albumin (g/dL)	0.64	0.26–1.51	0.32	–	–	–
Baseline C-reactive protein (mg/dL)	1.16	0.86–1.66	0.37	–	–	–
Baseline erythrocyte sedimentation rate (mm/h)	1.02	0.99–1.05	0.22	–	–	–
Week 0–14 exposure (AUC) goal, 79 348 μg h/mL	0.31	0.11–0.84	0.02	–	–	–

Abbreviations: AUC, area under concentration curve; CI, confidence interval; OR, odds ratio.

TABLE 3

Cox regression analysis of time to antibodies to infliximab (ATI) development

	HR	95% CI	*P* value
Age <10 years	1.86	0.99–3.49	0.05
Starting dose <7.5 mg/kg	2.09	0.98–4.45	0.05
Baseline Neutrophil CD64 activity ratio >6	2.55	1.14–5.70	0.02
Week 0–14 exposure (AUC) goal, 79 348 μg h/mL	0.48	0.27–0.84	0.009
HLA DQA*105	0.75	0.38–1.50	0.43

Abbreviations: AUC, area under concentration curve; CI, confidence interval; HR, hazard ratio.

TABLE 4

Antibodies to infliximab (ATI) resolution amongst patients with intervention (n = 32)

Infliximab optimisation	ATI resolved (n = 12)	ATI not resolved (n = 20)
Shortened interval (n = 9)	5	4
Dose escalation (n = 13)	3	10
Both interventions (n = 10)	4	6
Immunomodulator added (MTX, n = 3)[a]	–	3

Abbreviation: MTX, methotrexate.

[a]All three patients also had dose/interval changed.