scientific reports

Check for updates

OPEN

Establishment and internal validation of a model to predict the efficacy of Adalimumab in Crohn's disease

Fang Wang[1,3,4], He Zhou[1,4], Yujie Zhang[2], Yu Da[1], Tiantian Zhang[1], Yanting Shi[1], Tong Wu[1]✉ & Jie Liang[1]✉

Background: Clinically, the ability to distinguish which Crohn's disease patients can benefit from Adalimumab is limited. Aims: This study aimed to develop a model for predicting clinical remission probability for Crohn's disease patients with Adalimumab at 12 weeks. The model assists clinicians in identifying which Crohn's disease patients are likely to benefit from Adalimumab treatment before starting therapy, thus optimizing individualized treatment strategies. Methods: Demographic and clinical characteristics of Crohn's disease patients were utilized to develop a model for clinical remission probability. LASSO regression was used to select predictive factors, and predictions were made using a logistic regression model. The model was internally validated using the bootstrap method (resampling 1000 times). Results: 68 patients with Crohn's disease were enrolled in this study. Clinical remission was observed in 55.9% at 12 weeks. Three variables were selected through the least absolute shrinkage and selection operator regression method, including Adalimumab-positive cell count, disease duration, and neutrophil count of Crohn's disease patients. A predictive model was constructed by multivariate logistic regression (Adalimumab-positive cell count (OR, 1.143; 95%CI, 1.056–1.261), disease duration (OR, 0.967; 95%CI, 0.937–0.986), and neutrophil count (×10^9/L) (OR, 1.274; 95%CI,1.014–1.734)). The predictive model yielded an area under the curve of 0.866 (95%CI, 0.776–0.956), and in the internal validation, the area under the curve was 0.870 (95%CI, 0.770–0.940). Conclusions: This model provides a convenient tool to assess the likelihood of patient remission prior to Adalimumab treatment, thereby supporting the development of personalized treatment plans.

Keywords Crohn's disease, Prediction, Nomogram

Inflammatory bowel disease (IBD) is a chronic, idiopathic, and relapsing disease of the gastrointestinal tract, including ulcerative colitis (UC) and Crohn's disease (CD). The geographical epidemiology of IBD generally varies widely, and higher prevalence has traditionally been reported in Western countries. But in recent years, newly industrialized countries have rapidly reported increased incidence and prevalence of IBD, possibly due to industrialization, urbanization, culture, and westernization of diets[1,2]. In China, the number of patients has shown a rapid rise in the past 20 years, and it is expected that by 2050, the number of IBD patients in China will reach 1.5 million[3].

The use of biologics has dramatically improved the management of CD patients. Adalimumab (ADA) is a fully human-originated TNF-α antibody used subcutaneously. Thus, based on its safety and convenience, more and more IBD patients who meet the indications are using ADA. It is associated with a reduced risk of surgery and hospitalization. Both randomized controlled trials and real-world data demonstrate that ADA is safe and effective in treating CD[4–6].

However, approximately one-third of patients are primary non-responders, and 23-46% experience secondary response loss[7]. Thus, defining which patients will have no response and which factors are related is crucial. Factors affecting the efficacy of biologics are constantly being explored. Genetic variation may impact ADA

[1]State Key Laboratory of Holistic Integrative Management of Gastrointestinal Cancers, National Clinical Research Center for Digestive Diseases, Xijing Hospital of Digestive Diseases, Fourth Military Medical University, Xi'an, China. [2]Department of Histology and Embryology, School of Basic Medicine, Xi'an Medical University, Xi'an, China. [3]Department of Gastroenterology, Fenyang Hospital of Shanxi Province, Fenyang, China. [4]Fang Wang and He Zhou contributed equally to this work. ✉email: tongwu0725@fmmu.edu.cn; liangjie@fmmu.edu.cn

response. Some researchers reported that genetic markers could select the treatment for the individual patient[8]. In Danish patients with CD, three single nucleotide polymorphisms (SNPs) were associated with anti-TNFα (TNFRSF1A (rs4149570), IL18 (rs187238), and JAK2 (rs12343867)), the multi-SNP model predicted response rate of CD patients more than 82% [9]. Also, smoking[10], disease duration[11], C-reactive[11,12], previous anti-TNFα therapy[13] and previous surgery[10,11] may have a potential role in the prediction of the response. Moreover, a study that included 25 CD patients through local spraying of FITC-ADA with confocal molecular in vivo imaging technology found that the cell membrane surface expression of TNF-α had an excellent predictive effect on the efficacy of ADA treatment in CD patients. They found that the higher the numbers of TNF-α (>20), the higher the proportion of patients achieving clinical remission at 12 weeks. 92% of CD patients with TNF-α>20 achieve clinical remission with ADA treatment[14]. However, in vivo, fluorescence imaging has not been widely promoted in clinical practice.

Therefore, existing predictive models for the efficacy of Crohn's disease have limitations in accuracy and clinical application. This study developed a more practical predictive model by combining biomarkers and clinical data to address these shortcomings. In this study, we collected demographic and clinical characteristics of CD patients and stained immunofluorescence (IF) on biopsies of CD patients with FITC-ADA before ADA treatment. Build a new predicting model to predict the clinical remission probability of CD patients at 12 weeks.

Results

Patient characteristics

Between October 2020 and January 2023, 122 CD patients were registered in this study and accepted ADA treatment, and 54 of them were excluded from the primary cohort (including intestinal Behcet's disease, $n=5$; combined serious diseases, $n=2$; surgery, $n=2$; discontinuation of medication, $n=2$; absence of endoscopic biopsy tissue, $n=43$). 55.9% (38/68) of CD patients achieved clinical remission at 12 weeks (Fig. 1). Demographic and clinical characteristics of study participants are shown in Table 1. ADA-positive cell count (24.00 vs. 16.80, $p=0.001$), disease duration (17.90 vs. 42.40, $p=0.007$), and neutrophil count ($\times 10^9$/L) (6.21 vs. 4.50, $p=0.013$) between the groups of clinical remission and clinical non-remission were significantly different by student t-test. The rest of the variables had no significant differences.

Adalimumab-positive cell count

To clarify the efficacy of ADA-positive cell count in predicting the efficacy of ADA in treating CD, ADA-positive cell count between the clinical non-remission and clinical remission groups was compared, and the receiver operating characteristic (ROC) was plotted. As is shown in Fig. 2A-B, the ADA-positive cell count was increased in the clinical non-remission group compared to clinical remission (16.80 vs. 24.00 $p=0.001$). The areas under the ADA-positive cell count ROC curve (AUC) were 0.765 (95%CI, 0.641–0.890, Fig. 2C). The results collectively suggested that the number of ADA-positive cell counts can be one of the predictors of ADA treatment efficacy. However, more than the ADA-positive cell count is needed to predict effectiveness better. Therefore, more demographic and clinical characteristics before ADA treatment were collected for analysis, and a predictive model was constructed.

Prediction model establishment using selected factors

Variable selections were performed to develop a model for predicting clinical remission probability at 12 weeks of ADA treatment in CD patients. 3/28 variables collected from patients were selected based on non-zero coefficients calculated by LASSO regression analysis (Fig. 3A-B). These variables included ADA-positive cell count, disease duration, and neutrophil count ($\times 10^9$/L). Then, multivariable logistic regression analysis was performed based on the ADA-positive cell count (OR, 1.143; 95%CI, 1.056–1.261), disease duration (OR, 0.967; 95%CI, 0.937–0.986), and neutrophil count ($\times 10^9$/L) (OR, 1.274; 95%CI,1.014–1.734) to establish predictive model. The predictive model was as follows,

$$\text{Logit (P)} = -2.775 + 0.134 \times \text{ADA} - \text{positive cell count} - 0.035 \times \text{disease duration} + 0.243 \times \text{neutrophil count}$$

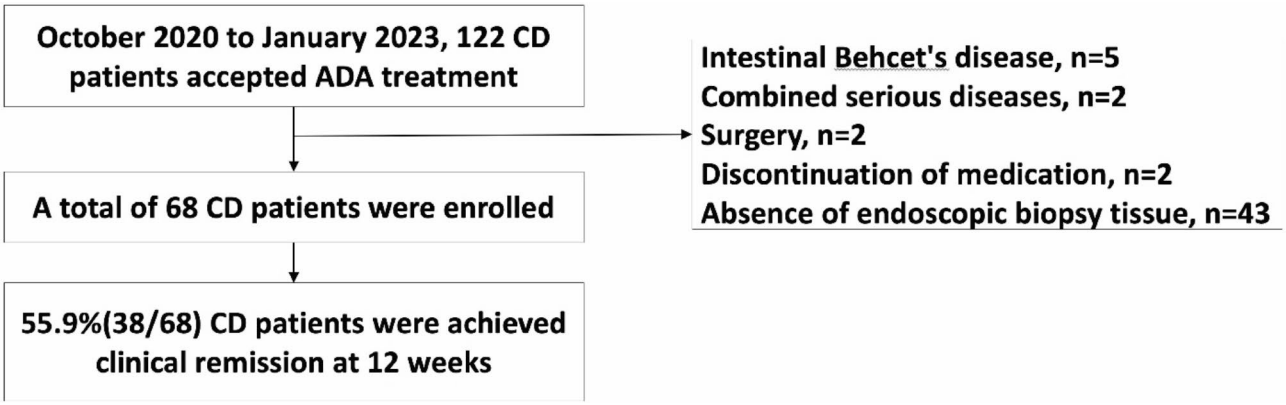

Fig. 1. Flow chart of CD patients in the study cohort.

	Clinical non-remission ($N=30$)	Clinical remission ($N=38$)	p
Male, n (%)	24 (80.0%)	24 (63.2%)	0.213
Age (years)	36.60 (13.2)	36.80 (15.5)	0.952
BMI	21.10 (3.14)	19.80 (3.04)	0.092
Smoking, n (%)	9 (30%)	8 (21.1%)	0.412
Disease duration (months)	42.40 (42.9)	17.90 (22.6)	0.007**
Age at diagnosis, n (%)			0.220
A1	8 (26.7%)	4 (10.5%)	
A2	14 (46.7%)	21 (55.3%)	
A3	8 (26.7%)	13 (34.2%)	
Disease location, n (%)			0.695
L1	6 (20.0%)	7 (18.4%)	
L2	12 (40.0%)	12 (31.6%)	
L3	12 (40.0%)	19 (50.0%)	
Disease behaviour, n (%)			0.468
B1	17 (56.7%)	27 (71.1%)	
B2	11 (36.7%)	10 (26.3%)	
B3	2 (6.67%)	1 (2.63%)	
HBI	8.27 (5.24)	7.00 (3.14)	0.248
SES-CD	13.20 (7.36)	13.90 (8.05)	0.715
History of medication			
5-Aminosalicylic acid, n (%)	22 (73.3%)	26 (68.4%)	0.862
Steroids, n (%)	13 (43.3%)	13 (34.2%)	0.605
Immunomodulatory, n (%)	6 (20.0%)	3 (7.89%)	0.169
Biologics, n (%)	6 (20.0%)	2 (5.26%)	0.126
ADA-positive cell count	16.80 (9.59)	24.00 (7.33)	0.001**
Leukocyte count ($\times10^9$/L)	6.95 (2.92)	7.00 (2.59)	0.942
Neutrophil count ($\times10^9$/L)	4.50 (1.84)	6.21 (3.57)	0.013*
Lymphocyte count ($\times10^9$/L)	1.74 (0.94)	1.59 (0.70)	0.140
NLR			
Haemoglobin (g/L)	130 (25.7)	125 (23.3)	0.396
Haematocrit	0.40 (0.07)	0.38 (0.07)	0.186
MCV (fl.)	87.20 (6.35)	85.00 (11.8)	0.331
MCHC (g/L)	320 (13.5)	324 (14.9)	0.368
Erythrocyte count ($\times10^{12}$/L)	4.60 (0.84)	4.40 (0.76)	0.174
Blood platelet count ($\times10^9$/L)	298 (113)	312 (118)	0.605
Albumin (g/L) C reactive protein (mg/L)	40.06 (8.15) 15.50 (24.7)	41.16 (6.02) 25.30 (34.4)	0.649 0.177
ESR (mm/hr)	30.60 (28.07)	28.10(21.91)	0.124

Table 1. Demographic and clinical characteristics of study participants. A1, ⩽16years; A2, 17–40years; A3, ⩾40years; L1, Terminal ileum; L2, Colon; L3, Ileum colon; B1, Non-narrow, non-fistula; B2, Narrow; B3, Fistula; E1, Proclitic; E2, Left-sided colitis; E3, Extensive colitis; NLR, Neutrophil-lymphocyte ratio; MCV, Mean corpuscular volume; MCHC, Mean corpuscular hemoglobin concentration; ESR, erythrocyte sedimentation rate; *, $p<0.05$; **, $p<0.01$

Then, a nomogram was constructed, which provided a convenient, personalized tool to predict the probability of clinical remission at 12 weeks among CD patients (Fig. 4). As using this nomogram, the total points of three variables for a particular CD patient would estimate the probability of remission at 12 weeks.

Predicting model accuracy and calibration

The AUC for the predictive model was 0.866 (95%CI, 0.776–0.956), and the sensitivity and specificity of the model are: 0.767 and 0.842. The internal validation using the bootstrap method (resampling = 1000) was 0.870 (95%CI, 0.770–0.940) (Fig. 5). The proposed model was well-calibrated (Fig. 6). The Hosmer-Lemeshow test yielded a non-significant p-value of 0.184, suggesting no statistical departure from a perfect fit between the predicted and observed values.

To assess its clinical validity, DCA was also performed. The DCA showed that when based on the nomogram in this study, the threshold probability of clinical remission in CD patients was 25–90% (Fig. 7), and application

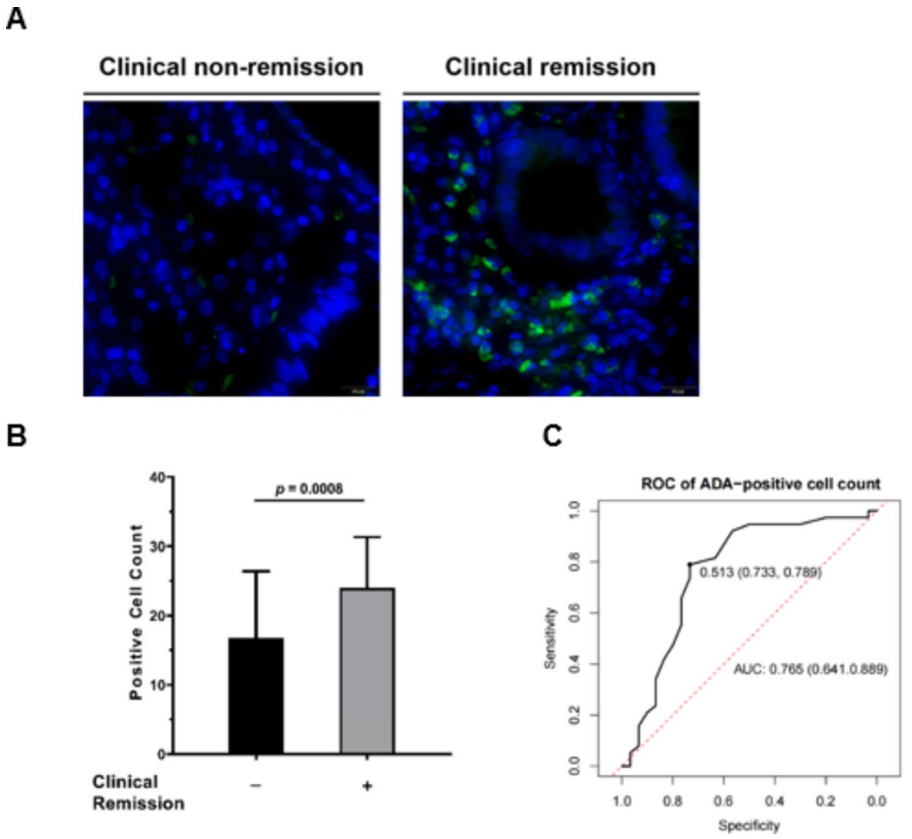

Fig. 2. ADA-positive cell count. (**A**) Selective immunofluorescence images of FITC-conjugated ADA-stained positive cells from the clinical non-remission and remission groups before ADA treatment. (**B**) The number of ADA-positive cells in the group of clinical non-remission and clinical remission (16.80 vs. 24.00 $p = 0.001$). (**C**) ROC curves of ADA-positive cell count in discriminating clinical remission from CD patients.

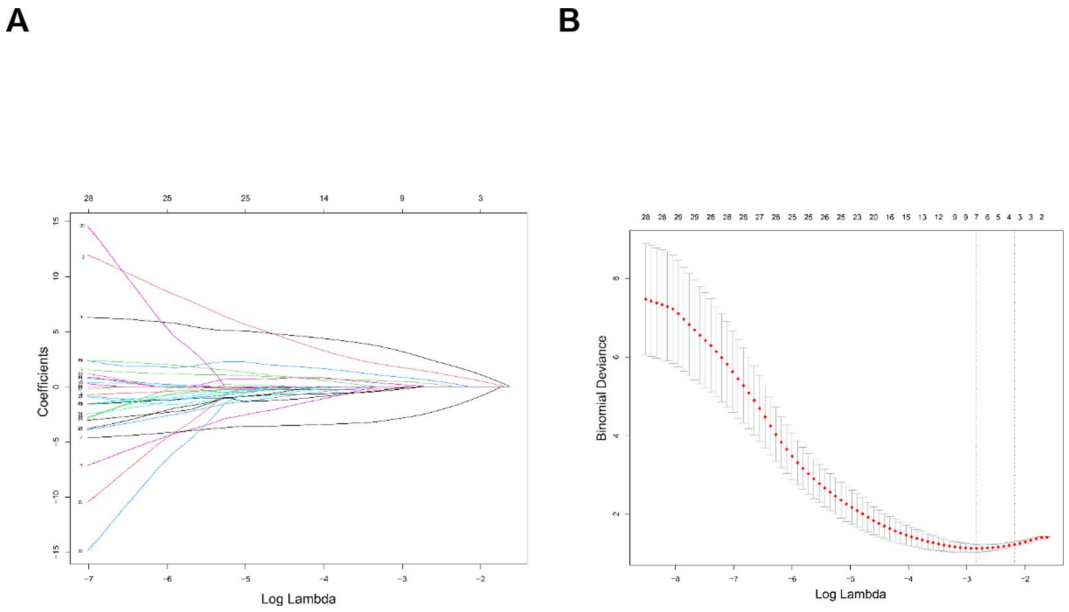

Fig. 3. Predictors were selected using LASSO regression analysis and tenfold cross-validation. (**A**) Selection of tuning parameter (lambda) for bias in LASSO regression. (**B**) Plot of the coefficients based on the log (lambda) series. This study chooses the predictor according to the 1-SE criterion (right dashed line), meaning three non-zero coefficients are chosen. LASSO, least absolute shrinkage and selection operator; SE, standard error.

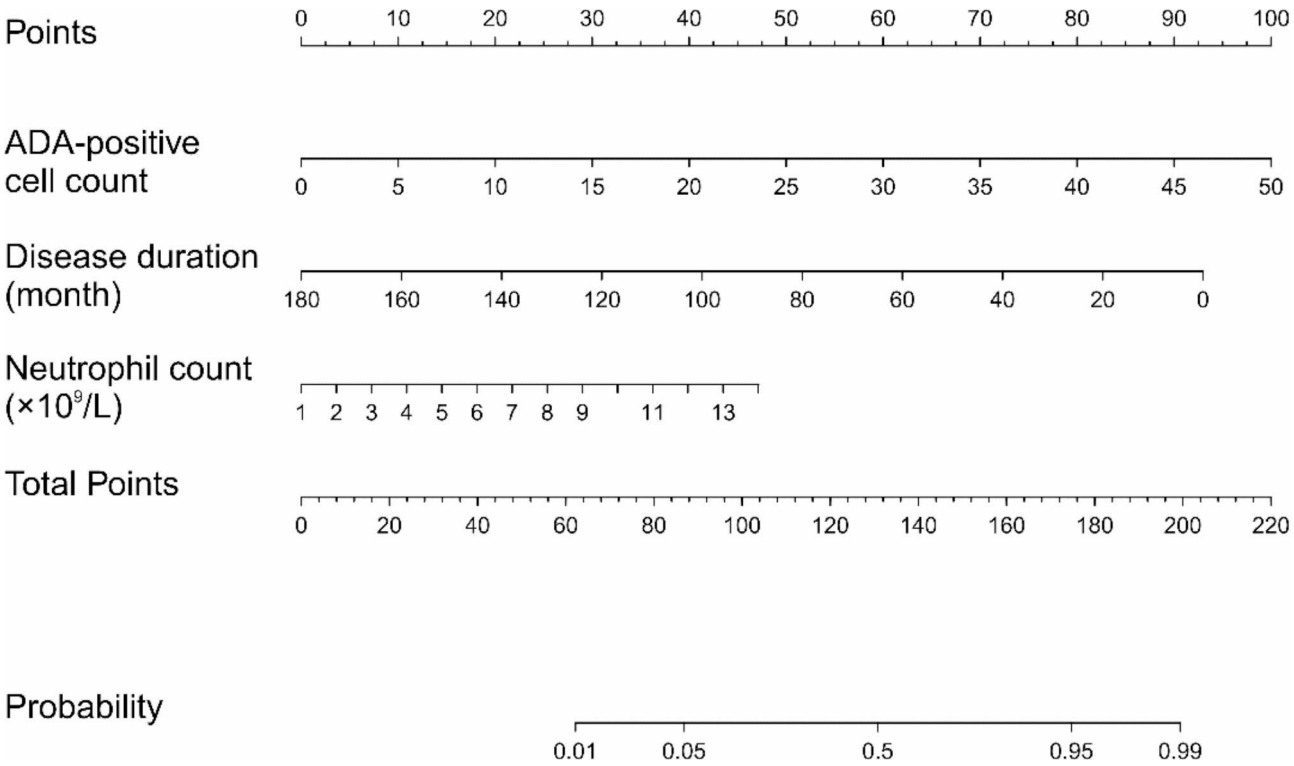

Fig. 4. Nomogram for predicting 12-week clinical remission and its algorithm. First, a point is found on the top rule for each variable for CD patients; then, all points are summed to collect the total points. Finally, find the corresponding predicted probability of 12-week clinical remission on the lowest rule.

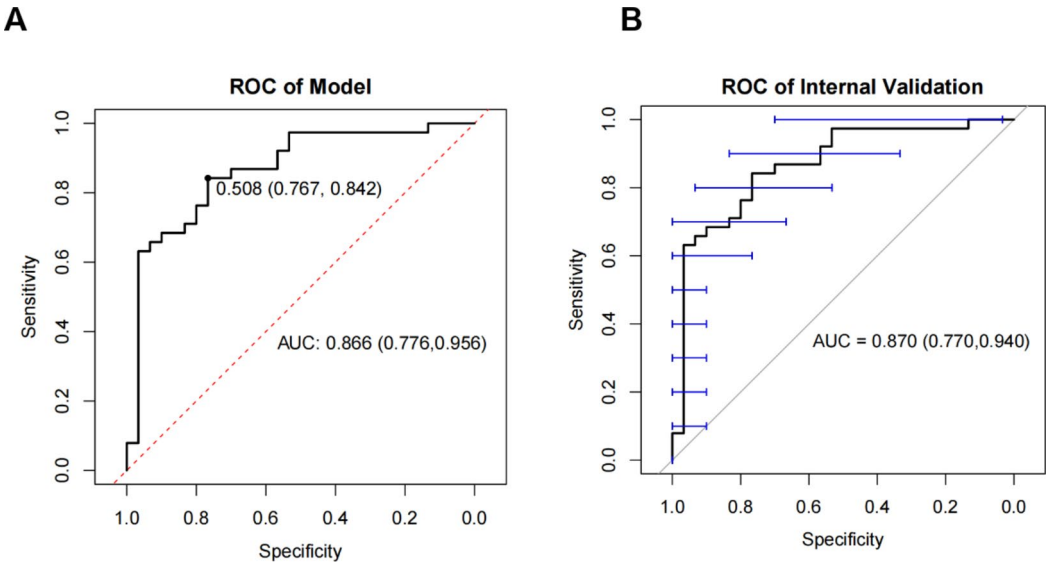

Fig. 5. The AUC (representing the discriminative power of the model) for model and internal validation. (**A**) indicates the AUC of the predictive model, and (**B**) indicates the AUC of the internal validation using bootstrapping (resampling = 1000). Blue lines represent 95% confidence intervals. The AUC is the area under the curve.

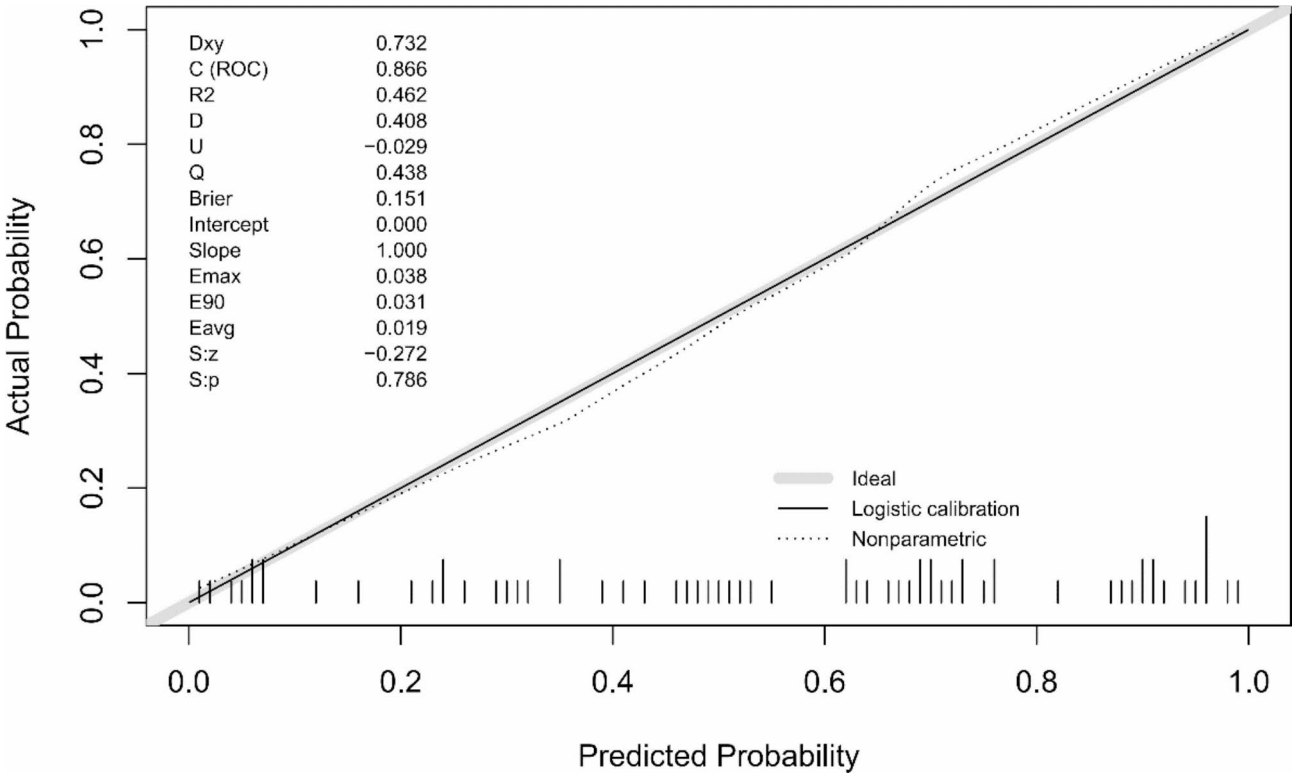

Fig. 6. The calibration curve of the predictive model shows the degree of agreement between the predicted and observed probability (Hosmer–Lemeshow test, $p > 0.05$, indicating goodness of fit). The solid line indicates the perfect prediction of the ideal model, and the dashed line indicates the model's performance.

of this nomogram to predict clinical remission at 12 weeks would add significantly more benefit than either the treat-all scheme or the treat-none scheme.

Discussion

Although the use of ADA has increased the cure rate of CD, not all patients benefit from it, including problems of non-response to treatment, serious side effects, and higher medication costs. A few medications would still be highly effective among all CD patients[8,15,16]. Therefore, providing a suitable individualized treatment strategy for patients among all available options is fundamental to solving the problem. Using the patient's phenotype and genetic characteristics to establish the most appropriate treatment for the patient might be a way to optimize the treatment plan and reduce the risk of adverse reactions and the cost of therapy[17].

Recent studies has shown a significant association between HLA-DQA1*05 carriers and loss of immunogenicity-mediated anti- TNFα responses [18]. But real-world research shows no correlation[20]. Our centre previously tested 20 patients with IBD and found that none of the 20 patients carried HLA-DQA1*05, but there were significant differences in the efficacy of TNFα treatment. Therefore, HLA-DQA1*05 was not performed in this study.

The cellular and molecular pathogenesis of inflammatory diseases, including UC, CD, ankylosing spondylitis (AS), rheumatoid arthritis (RA), and psoriasis, has continued to be revealed over the past 40 years[21]. Among all inflammatory cytokines and chemokines, TNFα was the first to be identified as an essential factor contributing to the inflammatory response process. As a result, anti-TNFα therapy was created and began to be used in the treatment of RA in the mid-1990s, and therapeutic goals started to evolve and move forward. Because of its potential role in disease remission and mucosal healing, anti-TNFα therapy reduces the risk of hospitalization, colectomy, and colorectal cancer, especially in patients with IBD. Mechanically, anti-TNFα mainly binds to TNFα in serum (s-TNFα) or on immune cell membranes (m-TNFα), leading to a critical role in reducing inflammatory responses[22,23]. A study, which included 25 CD patients, reported that m-TNFα was a good predictor of the short-term efficacy of anti-TNFα agents in treating CD patients. The greater the amount of TNFα on the surface of the cell membranes, the more patients achieved clinical remission at 12 weeks of treatment[14]. Another study has been conducted to characterize the expression of m-TNFα in CD patients before anti-TNFα therapy by immunohistochemical staining of intestinal biopsies. The results also showed that the high expression of m-TNFα was responsive to anti-TNFα treatment[24]. From our study, by in vitro immunofluorescence staining with FITC-ADA, we found that CD patients who reached clinical remission had a significantly higher number of ADA-positive cells than clinical non-remission before treatment. By plotting the ROC curve with an AUC of 0.765, although not as effective as in vivo fluorescence imaging reported in previous studies[14], it was still concluded that the number of ADA-positive cells was predictive of patient outcome.

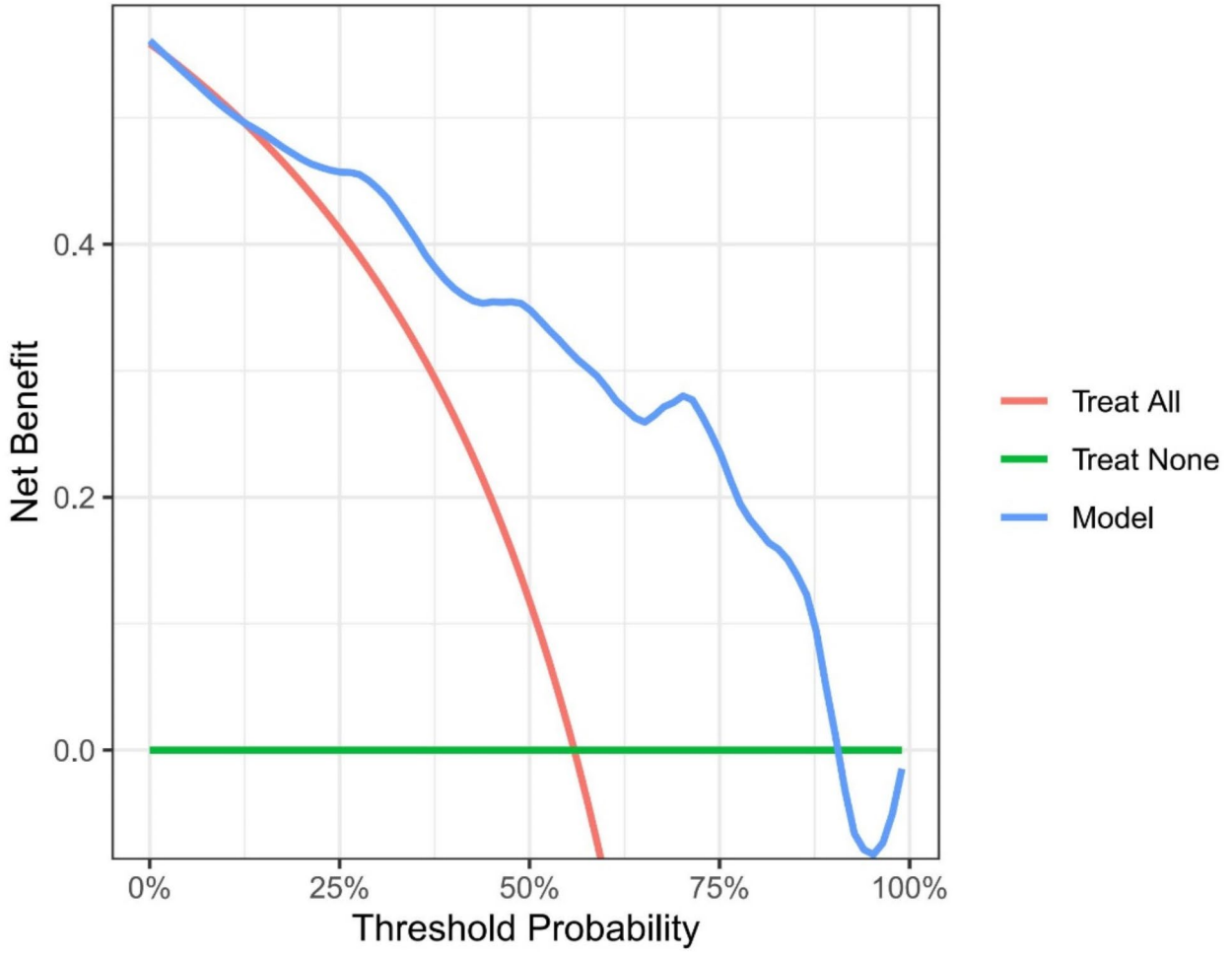

Fig. 7. DCA of the nomogram. The blue solid line represents the nomogram. The decision curve indicates that when the threshold probability of clinical remission at 12 weeks is between 25% and 90%, applying this nomogram would add a net benefit when compared with the treat-all or the treat-none strategies. DCA, decision curve analysis.

Subsequently, we selected the variables through LASSO regression and logistic regression constructed the model, and the three variables included in the model were ADA-positive cell count (OR, 1.143; 95%CI, 1.056–1.261), disease duration (OR, 0.967; 95%CI, 0.937–0.986), and neutrophil count ($\times 10^9$/L) (OR, 1.274; 95%CI,1.014–1.734). Therefore, we further investigated the clinical significance of each variable. For disease duration, some studies have concluded that the shorter the disease duration assessed, the better the response to early treatment in CD patients[25]. Post-hoc analyses of large clinical trials showed that patients with a disease duration shorter than two years had a higher chance of responding to anti-TNFα than patients with a longer disease duration. Thus, several studies also confirmed that CD patients with shorter disease duration respond better to anti- TNFα therapy, whether it is IFX[26,27], ADA[28,29], or certolizumab[30]. However, most studies could not confirm the existence of such an association in patients treated with anti-TNFα drugs[23,31,32]. Thus, our study concluded that the shorter the disease duration, the more influential the use of ADA.

Studies have not reported on the role of neutrophils in influencing anti-TNFα efficacy. However, the neutrophil-lymphocyte ratio (NLR) is believed to be a biomarker of the systemic inflammatory response[33]. It plays a role in the diagnosis and severity of paediatric IBD[34]. Nevertheless, in our research, it was unclear that NLR could be used as one of the prognostic indicators for ADA treatment. Collectively, our study found CD patients with high neutrophil count before ADA treatment, but the exact reason has not yet been reported.

As previously described, there is a continuous exploration of the factors related to the efficacy of anti-TNFα agents. Smoking[35] and prior use of anti-TNFα agents[36,37] have been identified as negative factors affecting the effectiveness of anti-TNFα agents in patients with CD. However, from our results, it can only be assumed that the number of smoking patients did not differ between the groups of clinical remission and clinical non-remission. Studies also have suggested that elevated C-reactive protein (CRP) levels are associated with the response to anti-TNFα agents in patients with CD[38]. CD patients with high baseline CRP levels responded more to anti-TNFα therapy than those with normal baseline CRP (90.8% vs. 82.6%; $p = 0.014$), and normalization of CRP levels early in treatment was associated with sustained long-term efficacy of anti-TNFα ($p < 0.001$). Consistently, our study

demonstrated that pre-treatment CRP was higher in CD patients with effective ADA treatment than in those with ineffective treatment.

In conclusion, the ADA-positive cell count obtained in vitro by immunofluorescence technique predicts the clinical efficacy of ADA treatment of CD patients for 12 weeks. Then, a nomogram was built to predict clinical remission probability at 12 weeks among CD patients. This nomogram incorporated three variables: ADA-positive cell count, disease duration, and neutrophil count. The nomogram showed good discriminatory ability, calibration, and clinical validity, providing an ideal method to predict ADA effectiveness and suggest anti-TNFα therapy selection in advance. It helps physicians stratify patients prior to treatment, optimizing individualized treatment decisions.

There are still limitations in our study. Our study did not confirm previously reported correlates of CRP, smoking, and albumin affecting ADA efficacy, possibly because most of the previously reported populations were from Western countries and may be different from our population. There are still shortcomings of a small sample size and the lack of external validation, suggesting that future multicenter studies are needed to further validate the model's applicability.

Methods

Study design and population

CD patients aged between 18 and 60 and treated with ADA for at least three months were enrolled in this study at the Xijing Hospital in China from October 2020 to January 2023. This study used both retrospective and prospective designs to recruit CD patients. CD patients who received or were scheduled to receive ADA voluntarily underwent a biopsy one week before the treatment. The patients were treated with ADA alone or with 5-aminosalicylic acid (5-ASA) or immunomodulators. The ADA was administered by subcutaneous injection, as 160 mg at week 0, 80 mg at week 2, 40 mg at week 4, and 40 mg every other week after week 4.

The following patients were excluded, as these conditions may affect the efficacy of Adalimumab and pose potential confounding factors. CD patients who were under-pregnancy, refused to undergo biopsy, combined with impaired coagulation function, and other diseases (moderate to severe heart failure, active tuberculosis, acute infection, or definite blood system diseases) were excluded from this study. Patients distinguished from the intestinal Behcet's disease with an ostomy, a total colectomy, or orally using steroids were not eligible either. Although 54 patients were excluded from the study, the selection bias is limited. We adhered to clear exclusion criteria, and a comparison of key characteristics showed no significant differences between excluded patients and those included in the analysis. This design aimed to minimize potential selection bias and ensure the reliability of the results.

All CD patients included in this study were confirmed and diagnosed through Clinical symptoms, endoscopy, and histological examination. Relative information on the demographic and clinical characteristics of CD patients was collected. The demographic characteristics included gender, age, body mass index (BMI), age at diagnosis, disease duration, disease behavior, disease location, previous IBD medication use, smoking history, and surgical history. The clinical characteristics included blood routine, stool routine, albumin, erythrocyte sedimentation rate (ESR), and C-reactive protein test results. The information obtained was all before ADA treatment. The study was reviewed and approved by the Medical Ethics Committee of the First Affiliated Hospital of the Air Force Medical University (KY20222333-C-1).

Biopsy collection

All samples were processed the same way. Biopsy was performed at the ulcer margin of CD patients. Necrotic tissue was avoided. Intestinal mucosal tissue of about 2 × 1 × 1 mm was clamped and fixed in 10% neutral-buffered formalin. After dehydrating with ethanol, biopsy tissues were embedded with paraffin as previously described[39].

Preparation of FITC-Adalimumab

FITC-conjugated ADA was synthesized per the manufacturer's protocol (ab102884, Abcam; Cambridge, UK), briefly as follows. The concentration of ADA was diluted to 4 mg/ml. Add 1 µl of modifier reagent to each 10 µl of ADA to be conjugated. Pipette the ADA sample with the added Modifier directly onto the lyophilized (FITC Mix). They were then incubated at room temperature for 3 h. The concentration of FITC-ADA was detected. Dilute the FITC-lgG complex with dialysate so that A280 < 2.0. Absorbance was measured at A280 and A492 to calculate FITC-ADA concentration. The binding molar ratio (F/P ratio) of FITC-ADA was calculated as follows: F/P molar ratio = A280 - (A492 × 0.35)/1.4 (reciprocal of the molar coefficient of the FITC-conjugated antibody). The final FITC-ADA concentration was 2 mg/ml.

Immunofluorescence staining

Paraffin-embedded tissue was 5 µm sectioned and cuffed for 1 h at 65 °C. Tissue sections were dewaxed, and the antigen was repaired for immunofluorescence staining. Incubation with Goat serum (#ZLI-9022, ZSGB-BIO; Beijing, China) for 30 min was used for blocking. The previously synthesized and verified FITC-ADA was used as the primary antibody and incubated at 4 °C overnight. After three times washing with PBST for 5 min, the slides were sealed using a quench-resistant sealer with DAPI (#36308ES20, YEASON; Shanghai, China). All slides were scanned using a laser scanning confocal microscope at 40× magnification (FV3000; Olympus), and five random fields of view were chosen for each slide. All positive cells in each field of view were counted, and the total average was recorded as the number of positive cells (ADA-positive cell count) for each slide.

Clinical outcomes

CD patients' severity of disease activity was assessed according to the simplified Crohn's Disease Activity Index score (Harvey Bradshaw Index, HBI)[40] within the week before the first treatment and after 12 weeks of

treatment. An HBI score of ≤ 4 was defined as remission, 5–7 as mild activity, 8–16 as moderate activity, and > 16 as severe activity. Clinical response was described as a decrease in HBI score of ≥ 3 points after treatment, and clinical remission was defined as a post-treatment HBI score of ≤ 4 points. Endoscopic scoring by Simplified Endoscopic Score for Crohn's Disease (SES-CD) to assess the condition of the patient's colonic mucosa as previously described[41].

Statistical analysis

GraphPad Prism (Version 8.3.0), SPSS (Version 26.0), and R (Version 4.2.0) software were applied for statistical analysis. Data conforming to normal distribution are expressed as mean ± SD. Statistical analyses were performed using the Student's t-test (Mann-Whitney test for non-normally distributed data). Rates were compared using the χ^2 test or Fisher's exact probability method. The least absolute shrinkage and selection operator (LASSO) regression technique was used for predictor selection. LASSO regression determined the optimal parameter λ through 10-fold cross-validation and selected variables with non-zero coefficients. Multivariable logistic regression analysis was used to develop a predictive model and a nomogram of clinical remission probability. The discriminatory capacity of the model was determined by calculating the area under the curve (AUC). The bootstrapping method (resampling = 1000) was employed for internal validation[42]. The reasons for choosing bootstrap over cross-validation include its good adaptability in small sample sizes and effective variance estimation, which allows for a more accurate assessment of model uncertainty. The calibration of the model was evaluated using the Hosmer-Lemeshow test, and the clinical validity of the model was assessed using decision curve analysis (DCA)[43]. $p < 0.05$ was considered statistically significant.

Data availability

The authors confirm that the data supporting the findings of this study are available within the article.

Received: 18 August 2024; Accepted: 9 December 2024
Published online: 15 January 2025

References

1. Kotze PG, et al. Progression of inflammatory bowel diseases throughout Latin America and the Caribbean: A systematic review. *Clin Gastroenterol Hepatol.* 2020;18(2):304-312.
2. Kaplan GG, Ng SC. Understanding and preventing the global increase of inflammatory bowel disease. *Gastroenterology.* 2017;152(2):313-321.e2.
3. Kaplan GG. The global burden of IBD: From 2015 to 2025. *Nat Rev Gastroenterol Hepatol.* 2015;12(12):720-727.
4. Hanauer SB, et al. Human anti-tumor necrosis factor monoclonal antibody (Adalimumab) in Crohn's disease: The CLASSIC-I trial. *Gastroenterology*. 2006;130(2):323-591.
5. Sohn IW, et al. Efficacy of Adalimumab in Korean patients with Crohn's disease. *Gut Liver*. 2016;10(2):255-261.
6. Moens A, et al. Translating results from VARSITY to real world: Adalimumab vs vedolizumab as first-line biological in moderate to severe IBD. *Inflamm Bowel Dis.* 2022;28(8):1135-1142.
7. Ben-Horin S, Kopylov U, Chowers Y. Optimizing anti-TNF treatments in inflammatory bowel disease. *Autoimmun Rev.* 2014;13(1):24-30.
8. Bek S, et al. Systematic review: genetic biomarkers associated with anti-TNF treatment response in inflammatory bowel diseases. *Aliment Pharmacol Ther*. 2016;44(6):554-567.
9. Bank S, et al. Polymorphisms in the NFkB, TNF-alpha, IL-1beta, and IL-18 pathways are associated with response to anti-TNF therapy in Danish patients with inflammatory bowel disease. *Aliment Pharmacol Ther.* 2019;49(7):890-903.
10. Moran GW, et al. Phenotypic features of Crohn's disease associated with failure of medical treatment. *Clin Gastroenterol Hepatol.* 2014;12(3):434-42.e1.
11. Narula N, et al. The efficacy and safety of either infliximab or Adalimumab in 362 patients with anti-TNF-α naïve Crohn's disease. *Aliment Pharmacol Ther.* 2016;44(2):170-180.
12. Peters CP, et al. Adalimumab for Crohn's disease: long-term sustained benefit in a population-based cohort of 438 patients. *J Crohns Colitis*. 2014;8(8):866-875.
13. Miyoshi J, et al. Early intervention with Adalimumab may contribute to favorable clinical efficacy in patients with Crohn's disease. *Digestion.* 2014;90(2):130-136.
14. Atreya R, et al. In vivo imaging using fluorescent antibodies to tumor necrosis factor predicts therapeutic response in Crohn's disease. *Nat Med.* 2014;20(3):313-318.
15. Barré A, Colombel JF, Ungaro R. Review article: Predictors of response to vedolizumab and ustekinumab in inflammatory bowel disease. *Aliment Pharmacol Ther.* 2018;47(7):896-905.
16. Stevens TW, et al. Systematic review: predictive biomarkers of therapeutic response in inflammatory bowel disease-personalised medicine in its infancy. *Aliment Pharmacol Ther.* 2018;48(11-12):1213-1231.
17. Gisbert JP, Chaparro M. Clinical usefulness of proteomics in inflammatory bowel disease: A comprehensive review. *J Crohns Colitis.* 2019;13(3):374-384.
18. Solitano V, et al. HLA-DQA1∗05 genotype and immunogenicity to tumor necrosis factor-α antagonists: A systematic review and meta-analysis. *Clin Gastroenterol Hepatol.* 2023;21(12):3019-3029.e5.
19. Sazonovs A, 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(1):189-199.
20. Pascual-Oliver A, et al. HLA-DQA1*05 was not associated with primary nonresponse or loss of response to first anti-TNF in real-world inflammatory bowel disease patients. *Inflamm Bowel Dis.* 2024;30(6):922-929.
21. Tracey D, et al. Tumor necrosis factor antagonist mechanisms of action: A comprehensive review. *Pharmacol Ther.* 2008;117(2):244-279.
22. Amini Kadijani A, et al. Transmembrane TNF-α density, but not soluble TNF-α level, is associated with primary response to infliximab in inflammatory bowel disease. *Clin Transl Gastroenterol.* 2017;8(9):e117.
23. Billiet T, et al. A matrix-based model predicts primary response to infliximab in Crohn's disease. *J Crohns Colitis.* 2015;9(12):1120-1126.
24. Vatansever A, et al. Effects of mucosal TNF-alpha levels on treatment response in Crohn's disease patients receiving anti-TNF treatment. *Hepatogastroenterology.* 2014;61(136):2277-2282.
25. Biancheri P, et al. The challenges of stratifying patients for trials in inflammatory bowel disease. *Trends Immunol.* 2013;34(11):564-571.

26. D'Haens G, et al. Early combined immunosuppression or conventional management in patients with newly diagnosed Crohn's disease: An open randomised trial. *Lancet*. 2008;371(9613):660-667.
27. Hyams J, et al. Induction and maintenance infliximab therapy for the treatment of moderate-to-severe Crohn's disease in children. *Gastroenterology*. 2007;132(3):863-1166.
28. Colombel JF, et al. Adalimumab for maintenance of clinical response and remission in patients with Crohn's disease: The CHARM trial. *Gastroenterology.* 2007;132(1):52-65.
29. Colombel JF, et al. Association between plasma concentrations of certolizumab pegol and endoscopic outcomes of patients with Crohn's disease. *Clin Gastroenterol Hepatol.* 2014;12(3):423-31.e1.
30. Schreiber S, et al. Increased response and remission rates in short-duration Crohn's disease with subcutaneous certolizumab pegol: An analysis of PRECiSE 2 randomized maintenance trial data. *Am J Gastroenterol.* 2010;105(7):1574-1582.
31. Sprakes MB, et al. Efficacy, tolerability, and predictors of response to infliximab therapy for Crohn's disease: A large single centre experience. *J Crohns Colitis.* 2012;6(2):143-153.
32. Barber GE, et al. Genetic markers predict primary non-response and durable response to anti-TNF biologic therapies in Crohn's disease. *Am J Gastroenterol. 2016;111(12):1816-1822.*
33. Langley BO, et al. Inflammatory bowel disease and neutrophil-lymphocyte ratio: A systematic scoping review. *J Clin Med.* 2021;10(18):4219.
34. Zahmatkesh A, et al. The role of platelet-to-lymphocyte ratio and neutrophil-to-lymphocyte ratio in the diagnosis and severity of inflammatory bowel disease in children. *Eur J Pediatr.* 2023;182(9):4263-4270.
35. To N, Gracie DJ, Ford AC. Systematic review with meta-analysis: the adverse effects of tobacco smoking on the natural history of Crohn's disease. *Aliment Pharmacol Ther.* 2016;43(5):549-561.
36. Gisbert JP, et al. Systematic review with meta-analysis: The efficacy of a second anti-TNF in patients with inflammatory bowel disease whose previous anti-TNF treatment has failed. *Aliment Pharmacol Ther.* 2015;41(7):613-623.
37. Billiet T, et al. Evolution of cytokines and inflammatory biomarkers during infliximab induction therapy and the impact of inflammatory burden on primary response in patients with Crohn's disease. *Scand J Gastroenterol.* 2017;52(10):1086-1092.
38. Jürgens M, et al. Levels of C-reactive protein are associated with response to infliximab therapy in patients with Crohn's disease. *Clin Gastroenterol Hepatol.* 2011;9(5):421-7.e1.
39. Dey P. Embedding of tissue in histopathology. *Basic and Advanced Laboratory Techniques in Histopathology and Cytology.* Springer Nature Singapore; 2022:29-34.
40. Harvey RF, Bradshaw JM. A simple index of Crohn's-disease activity. *Lancet.* 1980;1(8167):514.
41. Daperno M, et al. Development and validation of a new, simplified endoscopic activity score for Crohn's disease: The SES-CD. *Gastrointest Endosc.* 2004;60(4):505-512.
42. Collins GS, et al. Transparent reporting of a multivariable prediction model for individual prognosis or diagnosis (TRIPOD): The TRIPOD statement. *BMC Med.* 2015;13:1.
43. Van Calster B, et al. Reporting and interpreting decision curve analysis: A guide for investigators. *Eur Urol.* 2018;74(6):796-804.

Author contributions

JL and TW contributed to the study concept and design; FW and HZ organized the database. FW and HZ performed the immunofluorescence staining. FW, YZ, YD, and TZ performed the data analysis, statistical analysis, and interpretation. FW, HZ, and YS provided tables and pictures. All authors contributed to the manuscript's revision and read and approved the submitted.

Funding

Natural Science Foundation of Shaanxi Province, Key Industrial Innovation Project Fund (2023-ZDLSF-44); National Natural Science Foundation of China General Program (No. 82370588); Medical Personnel Training and Promotion Program (XJZT24QN41); National Natural Science Foundation of China Major Research Program Integration Project (No. 92259302); Independent Funds of the Key Laboratory (CBSKL2022ZZ34).

Declarations

Competing interests

The authors declare no competing interests.

Ethics approval statement

The study was conducted following the Declaration of Helsinki and approved by the Medical Ethics Committee of the First Affiliated Hospital of Air Force Medical University, Xi'an, China (KY20222333-C-1). The research was supported by the First Affiliated Hospital of Air Force Medical University.

Patient consent statement

All patients included in the study signed an informed consent.

Additional information

Correspondence and requests for materials should be addressed to T.W. or J.L.

Reprints and permissions information is available at www.nature.com/reprints.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.