Parenteral Methotrexate Is Efficient in the Treatment of Azathioprine Refractory Crohn's Disease

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ABSTRACT

Background: *There is limited data in the literature analyzing the efficacy of methotrexate in Crohn's disease used after thiopurine analogs. We aimed in our study to show the efficacy of methotrexate in Crohn's disease patients who failed to respond to thiopurine treatment.*

Methods: *The study included 29 azathioprine refractory patients with Crohn's disease. Intramuscular methotrexate (25 mg/week) in the induction of remission and intramuscular methotrexate (15 mg/week) in 29 CD patients with a median follow-up time of 13 months was performed. In 15 (51.7%) patients, methotrexate was used in combination with anti-Tumour necrosis factor (TNF) (combination group), while it was used in 14 (48.3%) patients in monotherapy (monotherapy group).*

Results: *The mean Harvey–Bradshaw index score significantly decreased in the follow-up period (Wk0 = 7.6, last visit = 4.5, P < .001). Remission and response rates at week 12 were 75.9% and 79.3%, respectively. Maintenance of remission (77.8% vs 37.5%, respectively, P = .1) and response rates (77.8% vs 50%, respectively, P = .3) due to last visit examination were numerically higher in combination group but they were not statistically significant. The cumulative probability of remission maintenance in patients with methotrexate therapy was 72.7%, 33.1%, and 22.0% at 1, 2 ,and 4 years after starting methotrexate, respectively.*

Conclusion: *Our results show that parenteral use of methotrexate is efficacious in inducing and maintaining remission as a step-up agent in azathioprine refractory Crohn's disease patients.*

Keywords: *Azathioprine, methotrexate, refractory Crohn's disease*

INTRODUCTION

Crohn's disease (CD) is a lifelong disease with relapses and remissions. Despite biologic agents having widespread use in clinical practice, immune modulators such as azathiopurine (AZA) and immunosupressants as metothrexate (Mtx) are still commonly used in corticosteroid dependent or refractory disease as an adjunctive therapy or as a steroid-sparing agent. Although Mtx monotherapy can be used either for remission induction or remission maintenance, AZA monotherapy can only be used for remission maintenance since the onset of its clinical response is delayed but it can be adopted as an adjunctive agent to induce remission.

Azathiopurine is one of the main treatment agents used to maintain long-term remission in patients in whom remission induction was obtained with corticosteroids (CS) or anti-TNFs. Although it is comparable with placebo to induce remission in active CD, they have been found to be efficient to decrease total CS dose and to prevent relapses in time when used in combination with CS or anti-TNFs.[1-3] However, approximately 20% of patients are resistant to AZA and also there are increased risks of lymphoma and non-melanoma skin cancer which raise physicians' fears and limit prescription of this medication on its long-term use.[4-6]

Since 1990s, alternatively to AZA, Mtx has been gradually adopted to manage refractory CD and was introduced to induce disease remission and to reduce the dosage of steroids in CD patients.[1] Although Mtx is regarded as an alternative treatment to AZA in refractory CD, there is limited data in the literature concerning the efficacy of Mtx as a step-up treatment agent for remission induction and maintenance of active CD. Therefore, we aimed to determine efficacy of parenteral Mtx treatment when used as a step-up therapeutic regimen instead of AZA in refractory CD.

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MATERIALS AND METHODS

Study Population

The study included AZA refractory patients with CD who were regularly followed up at the inflammatory bowel diseases (IBD) unit in a Turkish tertiary referral center hospital from January 2010 to December 2019. The inclusion criteria were: (1) age above 18 years, (2) active CD despite AZA monotherapy or AZA + anti-TNF combination therapy with a Harvey–Bradshaw activity index (HBI) >4.

Data Collection

The clinical data were retrospectively collected from the medical records of CD patients who were regularly followed up at IBD Unit of our center. The demographic data collected at baseline were gender, age, disease duration, disease location, and disease phenotype according to Montreal Classification.[7] Harvey–Bradshaw activity index scores and C-reactive protein (CRP) (mg/L) were collected at weeks 0, 12, 24, 52, and at last visit which is defined as the last visit performed in patients who were followed up more than 52 weeks.

Administration of Mtx Therapy

Mtx was administered intramuscular at a dose of 25 mg/week for 12 weeks (for induction of remission) followed by intramuscular Mtx 15 mg once weekly for maintenance of remission.

Outcome Measures

Remission was defined as a HBI equal or less than 4 at and treatment response was defined as at least 50% reduction in HBI levels. Remission and response rates were calculated at week 12 (Wk12), week 24 (Wk24), and week 52 (Wk52). Remission induction was defined as HBI ≤4 at Wk12. Maintenance of remission rates and treatment response rates at the end of the study were calculated by the last visit findings of the patients who were followed up more than 52 weeks.

Non-responder patients in the study were recommended to receive anti-TNF combination (in monotherapy group) or anti-TNF switch therapy (in combination group). In patients who rejected these therapies, 15 week systemic corticosteroid therapy (40 mg prednisolone at the beginning and tapered after 4th week) has been applied.

Statistical Analysis

The Statistical Package for Social Sciences (SPSS), version 17.0 software (SPSS Inc.; Chicago, IL, USA) was used for the statistical analysis. Descriptive statistics (frequencies, means, and standard deviations, median and minimum–maximum) were calculated. Categorical variables were provided as percentages. Comparisons of continuous variables were made using the Student's *t*-test or Mann–Whitney *U* test depending on normality of distribution. The chi-square test or Fisher's exact test (when chi-squared test assumptions do not hold due to low expected cell counts) was used to compare categorical variables in different groups. We utilized Friedman test to compare HBI and CRP values at baseline with those at follow-up (Wk12, Wk24, Wk52, and last visit). A Kaplan–Meier survival analysis was performed to predict relapse-free survival in patients who received Mtx therapy.

Ethics Approval for the Study

The study was approved by local the Ethics Committee of Türkiye Yüksek İhtisas Education and Research Hospital.

RESULTS

Study Population

Our study included 33 CD patients who were resistant to AZA therapy and received Mtx as a step-up therapy. Four patients were excluded from the study due to drug intolerance (1 patient, nausea and vomiting) or adverse events (3 patients, hepatotoxicity). Seventeen (58.6%) of the remained 29 patients who participated in the study were female. Mean age of the participants was 41.2. The median follow-up time of the patients after initiation of Mtx therapy was 13 months (min-max: 6-49). In 15 (51.7%) patients, Mtx was swapped with AZA who were using AZA + anti-TNF combination (combination group), while it was changed with AZA in 14 (48.3%) patients who were previously receiving AZA monotherapy (monotherapy group). Table 1 shows demographic and clinical characteristics of the study population.

Treatment Results

Mean HBI score and CRP were 7.6 (±SD = 2.3) and 20.7 (±SD = 17.9), respectively, at the beginning of the Mtx treatment, while they were 4.58 (±SD = 2.3) and 14.8 (±SD = 18.4) at 12th week (Wk12) of treatment, 4.5 (±SD = 2.2) and 11.1 (±SD = 13.2) at the last visit ($P < .001$ for HBI, $P = .08$ for CRP) (Figure 1). Remission and response rates at Wk12 were 75.9% and 79.3%, respectively. Maintenance of remission was achieved in 23/29 (79.3%) patients at 24th week (Wk24), 18/27 (66.7%) at 52nd week (Wk52), and 10/17 (58.8%) patients at the last visit. Response rates at Wk24, Wk 52, and last visit were 79.3%, 66.7%, and 64.7%, respectively (Figure 2).

Table 1. Demographic and Clinical Characteristics of the Study Population

	Monotherapy, n = 14	Combination, n = 15	*P*	Total
Age (mean ± SD) (years)	44.2 ± 12.5	38.3 ± 9.6	.1	41.2 ± 11.3
Gender (female) (n, %)	8 (57.1)	9 (60.0)	.8	17 (58.6)
Disease duration (median, min-max) (months)	79.5 (7-252)	88.0 (27-201)	.8	88.0 (7-252)
Follow-up time (median, min-max) (months)	13.0 (6-49)	13.0 (6-40)	.9	13.0 (6-49)
Disease localization				
Ileal (n, %)	5 (35.7)	4 (26.7)		9 (31.0)
Ileocolic (n, %)	6 (42.9)	6 (40.0)	.8	12 (41.4)
Colonic (n, %)	3 (21.4)	5 (33.3)		8 (27.6)
Disease phenotype				
Inflammatory (n, %)	5 (35.7)	6 (40.0)		11 (37.9)
Stricturing (n, %)	5 (35.7)	6 (40.0)	.8	11 (37.9)
Penetran (n, %)	4 (28.6)	3 (20.0)		7 (24.1)
Active smoking (n, %)	3 (21.4)	4 (26.7)	.7	7 (24.1)
Anti-TNF treatment IFX/ADA (n, %)		11 (73.3)/4 (26.7)		
Steroid experience				
Intolerance (n, %)	10 (71.4)	3 (20.0)		13 (44.8)
Steroid resistance (n, %)	4 (28.6)	10 (66.7)	.01	14 (48.3)
Steroid dependency (n, %)	-	2 (13.3)		2 (6.9)

SD, standard deviation; IFX, infliximab; ADA, adalimumab.

Comparison of Monotherapy and Combination Groups in terms of Remission and Response Rates

Remission rates were comparable between monotherapy and combination therapy groups for Wk12 (85.7% vs 66.7%, respectively, *P* = .6), Wk24 (71.4% vs 86.7%, respectively, *P* = .3), and Wk 52 (53.8% vs 78.6%, respectively, *P* = .2). Also response rates were not statistically different between groups for Wk12 (85.7% vs 73.3%, respectively, *P* = .3), Wk24 (71.4% vs. 86.7%, respectively, *P* = .3) ,and Wk 52 (53.8% vs 78.6%, respectively, *P* = .2). Also remission rates were comparable between monotherapy and combination therapy groups for second year (n = 3/8, 37.5% vs 77.8%, respectively, *P* = .1) and fourth year (n = 0/2, 0% vs n = 1/2, 50%, respectively, *P* = 1.0). Maintenance of remission (*P* = .1) and response rates (*P* = .3) due to last visit examination were numerically higher in combination group but they were not statistically significant (Figure 3).

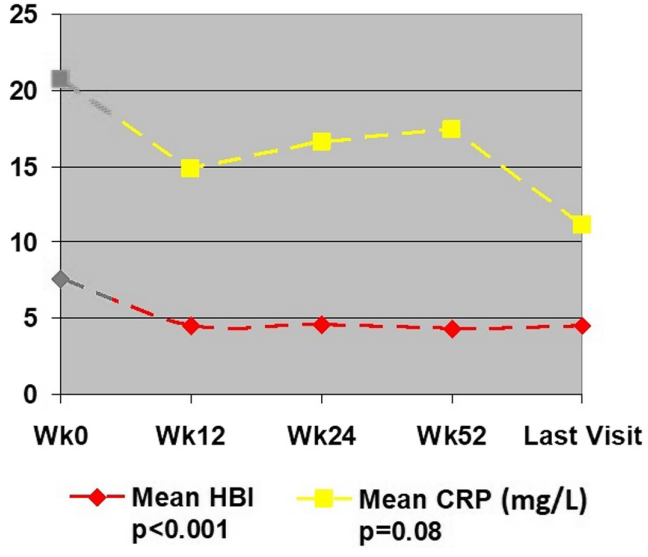

Figure 1. Changes in HBI and CRP values during metothrexate treatment. HBI, Harvey–Bradshaw index; CRP, C-reactive protein.

Comparison of Demographic and Clinical Characteristics of Patients in Terms of Remission Induction and Maintanence of Remission by the Last Visit

Basal median HBI value was significantly higher in patients without successful remission induction when compared with patients with successful remission induction (11.0 vs 6.0, *P* < .001) (Table 2). Also Table 3 shows the comparison of clinical and demographic characteristics of

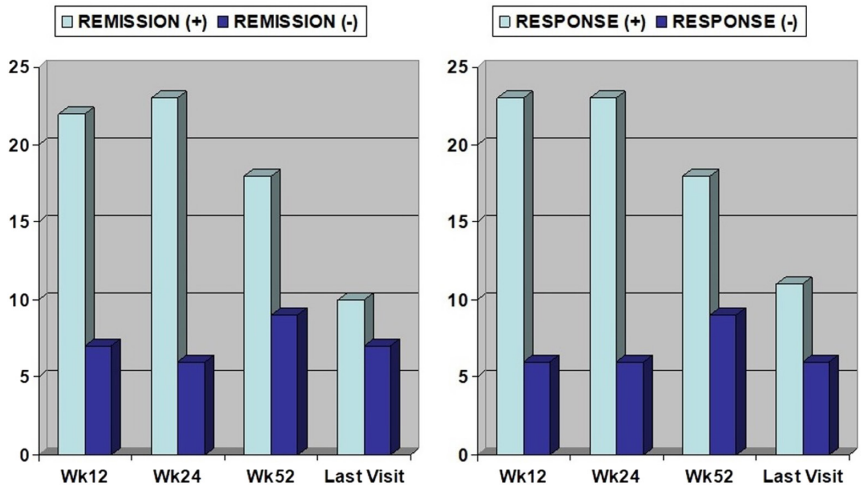

Figure 2. Remission and response rates of metothrexate treatment according to the treatment week.

patients with and without maintained remission by the last visit.

Cumulative Probability of Maintenance of Remission

The cumulative probability of remission maintenance in patients with Mtx therapy was 72.7%, 33.1%, and 22.0% at 1, 2, and 4 years after starting Mtx, respectively (Figure 4).

Safety

Metothrexate was well tolerated in patients who continued the study. No serious adverse event was reported during study period.

DISCUSSION

Our results documented the efficacy of parenteral Mtx treatment in the induction and remission of AZA

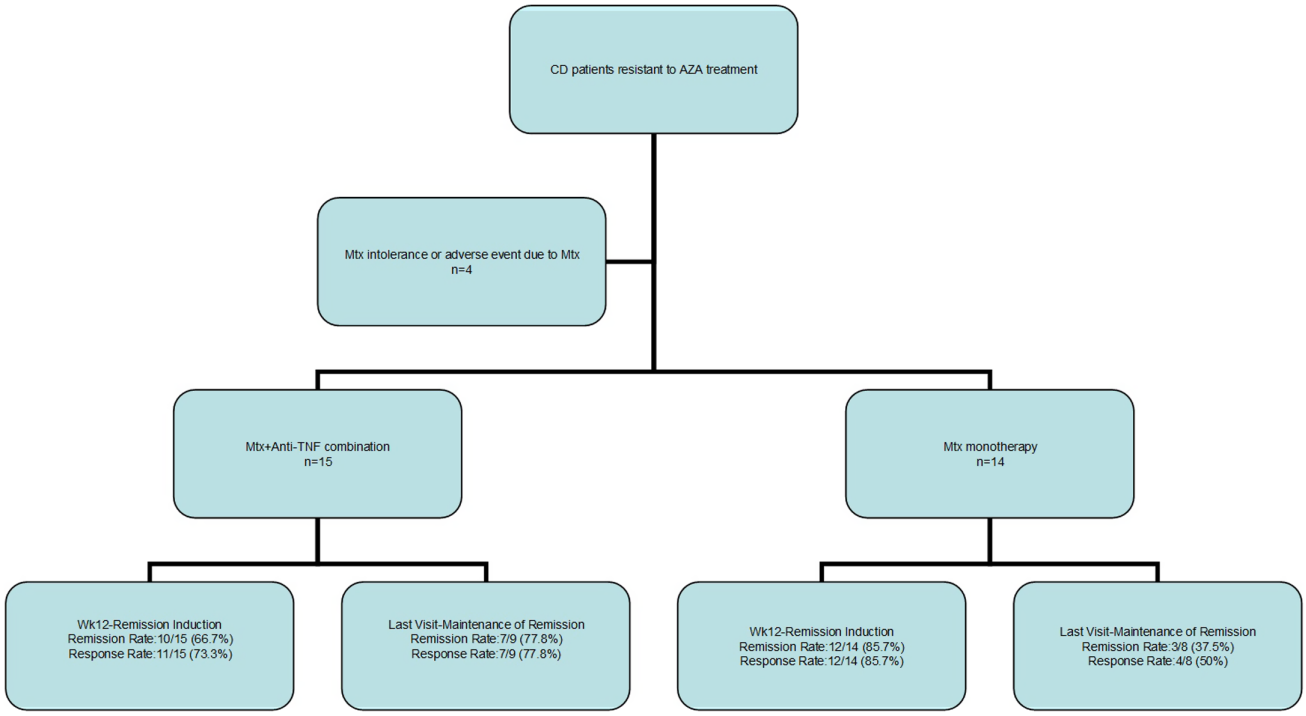

Figure 3. Outcomes of metothrexate treatment according to treatment groups.

Table 2. Comparison of Demographic and Clinical Characteristics of Patients in Terms of Successful Remission Induction

	SRI (+), N = 22 (75.9%)	SRI (–), N = 7 (24.1%)	*P*
Age (mean ± SD) (years)	42.0 ± 11.8	38.4 ± 9.8	.4
Gender (female) (n, %)	11 (50.0)	1 (14.3)	.1
Disease duration (median, min-max) (months)	115.0 (7-252)	72 (27-104)	.1
Follow-up time (median, min-max) (months)	12.5 (6-49)	14.0 (6-32)	.1
Basal HBI (mean ± SD)	6.6 ± 1.0	10.5 ± 2.7	<.001
Basal CRP (mg/L) (median, min-max)	20.3 (0.30-63.0)	28.0 (4.3-59.0)	.1
Disease localization			
Ileal (n, %)	7 (31.8)	2 (28.6)	
Ileocolic (n, %)	11 (50.0)	1 (14.3)	.1
Colonic (n, %)	4 (18.2)	4 (57.1)	
Disease phenotype			
Inflammatory disease (n, %)	9 (40.9)	2 (28.6)	
Stricturing disease (n, %)	10 (45.5)	1 (14.3)	.07
Penetrating disease (n, %)	3 (13.6)	4 (57.1)	
Active smoking (n, %)	6 (27.5)	1 (14.3)	.4
Steroid experience			
Intolerance (n, %)	10 (45.5)	3 (42.9)	
Steroid resistance (n, %)	11 (50.0)	3 (42.9)	.6
Steroid dependency (n, %)	1 (4.5)	1 (14.2)	

SD, standard deviation; CRP, C-reactive protein; SRI, successful remission induction; HBI, Harvey–Bradshaw index.

refractory CD. Although currently there is a tendency of widespread use of biologic agents and immunomodulator agents in earlier phases of CD, Mtx is usually considered as a second choice drug in patients who are intolerant to AZA treatment and has a limited use in IBD practice. A major reason for this seems to be concerns about potential hepatotoxicity of Mtx.[8] Despite this concern, in our study, only 4 patients could not continue remission induction due to Mtx intolerance or hepatotoxicity.

Efficacy of Mtx treatment in CD treatment was documented in a Cochrane database systematic literature review. According to this review, parenteral (intramusculer) Mtx at a dosage of 25 mg/week has been shown to be beneficial for induction of remission in patients who were unable to wean steroid therapy and complete withdrawal of steroids. Also, intramuscular Mtx at a dosage of 15 mg/week was superior to placebo for maintenance of remission in CD. This review documented that oral Mtx at a dosage of 12.5 mg/week was not superior to placebo in terms of maintenance of remission.[9] In another study, bioavailability of oral versus subcutaneous low-dose methotrexate in patients with CD has been compared and it has been found that oral bioavailability of Mtx is reduced in patients with severe intestinal disease.[10] All these findings suggest that the dosage and route of administration may influence the success rate of Mtx therapy in CD. In our study, Mtx was administered intramuscular at a dose of 25 mg/week for 12 weeks (for induction of remission) followed by intramuscular Mtx 15 mg once weekly for maintenance of remission as routinely used in our IBD department.

In the literature, studies comparing efficacy of Mtx and thiopurines in CD show that the efficacy of these 2 agents are comparable. In the study by Ardizzone et al[11] Mtx has been shown to achieve numerically but not statistically higher remission rates than AZA after 3 months of treatment in patients with CD. Also, in each treatment group, more than 50% of patients were in remission at 6 months. In another study, Maté-Jiménez et al[12] found that induction of remission (93.3% vs 80%, respectively) and maintaining remission (53.3% vs 66.6%, respectively) rates of thiopurines and Mtx were similar in steroid refractory CD patients.

Table 3. Comparison of Demographic and Clinical Characteristics of Patients in Terms of Maintenance of Remission by the Last Visit

	MR (+), n = 10 (58.8%)	MR (–), n = 7 (41.2%)	*P*
Age (mean ± SD) (years)	41.5 ± 12.1	45.0 ± 6.7	.5
Gender (female) (n, %)	3 (30.0)	2 (28.6)	1.0
Disease duration (median, min-max) (months)	56.5 (7-198)	104.0 (48-144)	.2
Follow-up time (median, min-max) (months)	14.0 (13-40)	32 (13.0-49)	.08
Basal HBI (mean ± SD)	8.6 ± 2.8	7.2 ± 1.7	.2
Basal CRP (mg/L) (median, min-max)	22.3 (1.6-63.0)	20.0 (5.9-45.0)	.9
Disease localization			
Ileal (n, %)	3 (30.0)	3 (42.8)	
Ileocolic (n, %)	2 (20.0)	2 (28.6)	.6
Colonic (n, %)	5 (50.0)	2 (28.6)	
Disease phenotype			
Inflammatory disease (n, %)	4 (40.0)	2 (28.6)	
Stricturing disease (n, %)	5 (50.0)	1 (14.3)	.1
Penetrating disease (n, %)	1 (10.0)	4 (57.1)	
Active smoking (n, %)	2 (20.0)	2 (28.6)	1.0
Steroid experience			
Intolerance (n, %)	3 (30.0)	4 (57.1)	
Steroid resistance (n, %)	6 (60.0)	2 (28.6)	.5
Steroid dependency (n, %)	1 (10.0)	1 (14.3)	

SD, standard deviation; CRP, C-reactive protein; MR: maintenance of remission; HBI, Harvey–Bradshaw index.

Studies analyzing efficacy of Mtx in CD as a second-line agent after AZA therapy is usually performed in patients who are intolerant or refractory to AZA treatment. In a retrospective study, Mtx was administered to AZA refractory patients in addition to AZA intolerant patients and remission induction was achieved in 72% of patients with intramuscular 25 mg/week Mtx. Also, after induction therapy, Mtx was given orally and the probability of relapse was found to be 78% at weeks 50.[13] In another retrospective study, Mtx was administered in 22 steroid-dependent CD patients ([55%] were intolerant to thiopurines and in the remaining, 10 [45%] were azathioprine non-responder). All patients received 25 mg/wk parenterally (subcutaneous or intramuscular) Mtx for a total 16 weeks. At the end of 16 weeks, treatment response rate was 77%.[14] In our study, although all patients were only refractory to AZA treatment, with a intramuscular administration of Mtx at a 25 mg/week dosage, remission induction was obtained in 75.9% of patients.

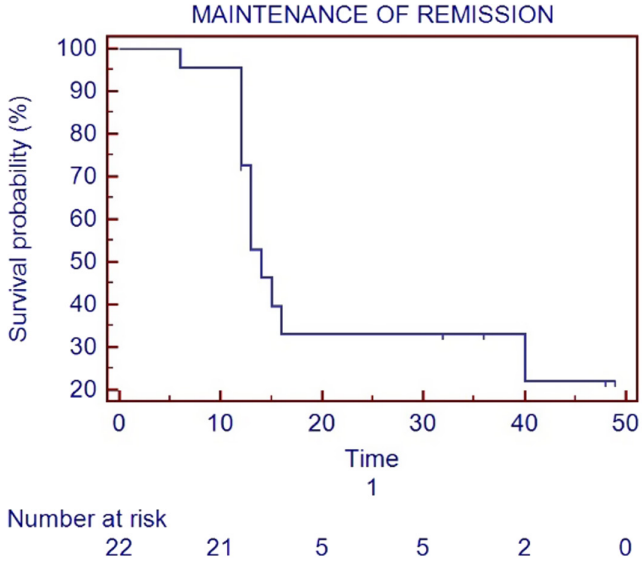

Figure 4. Kaplan–Meier curve of maintenance of remission probability after remission for patients on metothrexate therapy.

Long-term studies show that, although Mtx treatment has satisfactory response and remission rates for the first year of the treatment, up to 50%-75% of patients lose their initial response after 3 years of treatment.[13-16] In the study by Din et al[13] the cumulative probability of relapse was 78% at weeks 50 of Mtx therapy. In another retrospective

study, remission rates were 72%, 46%, and 39% at 1, 2, and 3 years after starting Mtx.[14] Charpignon et al[17] concluded in their study that, dose-tapering beyond the first 3 months of treatment could be one of the causes of loss of efficacy of Mtx. Similar to these findings, in our study, cumulative probability of relapse rates were 27.3%, 66.9%, and 78% at 1, 2, and 4 years.

In our study, response and remission rates in those patients treated with or without a concomitant anti-TNF agent were similar. However, maintenance of remission and response rates due to last visit examination were numerically higher in combination group. Due to its retrospective design, it is difficult to make exact conclusions about efficacy of concomitant use of Mtx and anti-TNF agents in our study. Another limitation of our study for this issue is the lack of measurement of anti-TNF trough levels and anti drug antibody levels.

Another limitation of our study is the absence of mucosal healing rates. We could not analyze mucosal healing data in our study since none of the patients had objective endoscopic scoring as Simple Endoscopic Score for CD in their medical records. Also, in the current literature, there is extremely rare data on the effects of MTX on mucosal healing. Only 1 prospective study assessed the mucosal healing rate in Mtx-treated patients and only 2 of the 18 patients achieved mucosal healing.[18] In a more recent retrospective study, Huang et al[6] showed that mucosal healing rate was 47.4% (9/19) after 16-week treatment of Mtx in steroid-dependent or refractory patients.[6]

Although it has some limitations, due to our best knowledge, our study is unique in the literature analyzing the efficacy of Mtx therapy in AZA refractory patients. By excluding AZA intolerant patients, we aimed to show the efficacy of Mtx therapy when used as a step-up treatment agent. Our results show that parenteral use of Mtx in CD has acceptable and satisfactory clinical remission and response rates when used in AZA refractory patients. In our opinion, these findings also suggest that parenteral Mtx should be considered in steroid refractory CD as a primary choice rather than as a substitute of AZA. Current data about efficacy of Mtx in CD is provided generally via retrospective small sample size studies. Further large size randomized studies are required to document the efficacy and safety of Mtx in CD.

Ethics Committee Approval: The study was conducted according to the ethical standards specified in the 1964 Helsinki Declaration. In our study, research and publication ethical rules were followed. The study was approved by local the Ethics Committee of Türkiye Yüksek İhtisas Education and Research Hospital.

Informed Consent: N/A.

Peer-review: Externally peer-reviewed.

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