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Adalimumab Monotherapy and a Combination with Azathioprine for Crohn's Disease: A Prospective, Randomized Trial

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Abstract

Background and Aims: The efficacy of azathioprine for Crohn's disease under adalimumab treatment remains obscure.

Methods: In an open-labelled prospective study, we evaluated the efficacy of adalimumab with and without azathioprine in patients with active Crohn's disease, who were naïve to biologics and thiopurines. The patients were randomly assigned to subcutaneous administration of adalimumab [monotherapy group] or to exactly the same schedule of adalimumab with azathioprine [25–100 mg daily] [combination group] for 52 Weeks. The primary endpoint was clinical remission at WWeek 26. We also evaluated the score for simple endoscopic severity of Crohn's disease before the therapy and at WWeeks 26 and 52.

Results: A total of 176 patients were randomized to either the monotherapy group [*n* = 85] or to the combination group [*n* = 91]. Eighteen patients [21.2%] from the monotherapy group and 7 patients [7.7%] from the combination group withdrew owing to active disease, and 15 patients [16.5%] from the combination group and 1 patient [1.2%] from the monotherapy group withdrew due to side effects of the medications. Non-responder imputation analysis revealed that the remission rate at WWeek 26 did not differ between the monotherapy group and the combination group [71.8% vs 68.1%; OR 0.84, *p* = 0.63]. The rate of endoscopic improvement at WWeek 26 was significantly higher in the combination group [84.2%, *n* = 57] than in the monotherapy group [63.8%, *n* = 58] [*p* = 0.019].
Conclusion: The clinical efficacy of a combination of adalimumab and azathioprine at WWeek 26 did not differ from that of adalimumab monotherapy in patients with Crohn's disease naïve to both medications.

Key Words: Crohn's disease; anti-TNF-alpha antibody; immunomodulator

1. Introduction

A large number of clinical observations have confirmed that biologics, especially those developed against tumour necrosis factor alpha [TNF-α], are efficacious for the treatment of Crohn's disease [CD]. Among various anti-TNF-α monoclonal antibodies, infliximab [IFX] and adalimumab [ADA] are unequivocally effective for the induction and maintenance of remission for patients with active CD. It has also been shown that the therapeutic effect of IFX, a chimeric monoclonal antibody to TNF-α, is enhanced by simultaneous use of immunomodulators [IMs] such as azathioprine [AZA] and 6-mercaptopurine [6-MP]. In a prospective, randomized clinical trial, Colombel *et al*. showed that a combination of IFX and AZA was more efficacious than either medication for maintaining the remission of CD.[1]

ADA, a humanized monoclonal antibody, has been shown to be efficacious for the treatment of Western and Eastern patients with CD.[2,3] However, the benefit of the use of IMs in addition to ADA is a matter of debate.[4] In a sub-analysis of a prospective clinical trial of ADA, Colombel *et al*. failed to show any benefit of IMs for the maintenance of remission.[2] Other retrospective analyses from various countries did not confirm the efficacy of IMs for the patients treated with ADA.[5,6] In contrast, there have been single- and multi-centre cohort studies showing the efficacy of IMs for the treatment of CD treated with ADA.[7–9]

To date, there has been no prospective clinical trial investigating the effect of simultaneous IM administration to patients with CD treated with ADA. The still controversial effect of IM administration on the efficacy of ADA seems to be attributable to the lack of reliable evidence obtained by prospective evaluation. We thus conducted a multicentre, randomized, prospective, open-labelled trial to clarify this issue. This study was referred to as Deep Remission of Immunomodulator and Adalimumab Combination Therapy for Crohn's Disease [DIAMOND].

2. Methods

2.1. Patients

DIAMOND was a multicentre, randomized, prospective, open-labelled study. Patients with a diagnosis of moderate to severe CD, who were naïve to anti-TNF-α antibodies and IMs, were enrolled in the trial. The diagnosis of CD was based on the criteria determined by the Japanese Ministry of Health, Labour and Welfare.

The patients were males and females with CD of at least 3 months' duration, and their age ranged from 15 to 65 years. Moderate to severe CD was regarded as a disease with the Crohn's disease activity index [CDAI] ≥ 220.[10] The exclusion criteria were as follows; 1] patients with a contraindication for anti-TNF-α [severe infection, active mycobacterial infection, a past or present history of demyelinating disease, or clinically evident congestive heart failure]; 2] patients with a contraindication for thiopurines [peripheral white blood cell count of less than 3000/ml, or established possible pregnancy]; 3] patients with ongoing breastfeeding; 4] patients from whom informed consent could not be obtained; 5] patients with a previous history of anti-TNF-α use; 6] patients with a previous history of IMs [AZA, 6MP, methotrexate, tacrolimus, or cyclosporine] use; 7] patients with a malignant neoplasm; 8] patients with an interval of less than 6 months after the latest surgery; 9] patients with short bowel syndrome; 10] patients with ileostomy or colostomy; and 11] patients regarded as being inappropriate by the attending physician.

The protocol of the clinical trial was approved by the IRB at each institution and registered publicly on the UMIN registration [No. 000005146]. Informed consent to the study was obtained from each participant before inclusion. All the authors had access to the study data and reviewed and approved the final manuscript.

2.2. Study protocol

Each patient was treated by subcutaneous administrations of ADA [Humira] at doses of 160 mg at Week 0, 80 mg at Week 2, and thereafter 40 mg at every other week up to 52 weeks. The patients who were assigned to a combination of ADA and AZA [combination group] were further treated with oral administration of AZA during the investigation period, whereas those assigned to monotherapy with ADA [monotherapy group] were not administered AZA. Since DIAMOND was an open-labelled study, the patients in the monotherapy group were not treated with a placebo either. The patients in the combination group were initially treated with 25 mg or 50 mg/day of AZA and the dose was allowed to be increased to a maximum of 100 mg during the initial four weeks, under careful observation. The maximum dose of AZA was chosen based on erythrocyte 6-thioguanine nucleotide [6TGN] concentrations in Japanese patients with inflammatory bowel disease.[11] Follow-up data collections were conducted several weeks after a patient completed the study at Week 52 or immediately after withdrawal from the clinical trial.

Randomization was done centrally at the Clinical Research Centre in Keio University with the use of an adaptive randomization procedure. The items for stratification included the institution and the duration of CD. During the investigation period, oral mesalazine or sulphasalazine was maintained at a stable dose. Each physician was permitted to decrease the dose of systemic corticosteroids from that at entry. However, dose escalation of corticosteroids was prohibited.

2.3. Efficacy and safety

Scores of the CDAI were determined at Weeks 0, 2, 4, 12, 26 and 52. Ileocolonoscopy was done at the baseline, at Week 26 and at Week 52. The mucosal lesion at each colonoscopy was assessed according to the simple endoscopic score for CD [SES-CD].[12] The endoscopic images at the sites of mucosal involvement were recorded as still images and SES-CD at each colonoscopy was calculated by the attending physician. Clinical remission was defined as a CDAI score of less than 150 points.[10] A clinical response was defined as a reduction of CDAI from the baseline value by more than 70. Endoscopic responses at Week 26 and Week 52 were defined as a decrease of SES-CD of at least 8 points from the baseline, or SES-CD ≤ 4.[13]

Blood samples were collected from patients in the combination group, at Week 12, and processed to measurement of 6-TGN in red blood cells [RBCs]. Whole blood samples were collected in heparinized tubs and centrifuged. After removing plasma, RBCs were hydrolyzed with acid and extracted with phenylmercuric acetate/ethyl acetate. 6-TGN levels were measured by high-performance liquid chromatography.[14] We also collected serum samples from the patients in both groups at Week 26 and measured trough levels of ADA and anti-antibodies to ADA [AAA].[15,16] Based on the manufacturer's recommendations, serum AAA level of ≥ 12 μg/ml was regarded as positive for the antibody.[16]

2.4. Primary and secondary endpoints

On the basis of the results of SONIC study,[1] which compared treatments with IFX, AZA and a combination of the two, we designed DIAMOND as a superior study. The primary efficacy endpoint was the rate of clinical remission at Week 26. The rates of clinical remission at the other time points and the rate of a clinical response at each time point were the secondary efficacy endpoints. The secondary efficacy endpoints also included the rates of mucosal improvement at Weeks 26 and 52. The safety endpoint was the rate of any adverse events that occurred during the study period. We also assessed the rate of loss of response to ADA as a safety endpoint.

2.5. Statistical analyses

Analyses were undertaken using IBM SPSS Statistics 20 [IBM]. For the primary endpoint, namely the rate of clinical remission at Week 26, it was estimated that 200 patients would be needed to attain a power of 80% in order to detect a difference in remission rates of 20% between the combination and monotherapy groups. The calculation was based on the assumption that the rates of remission would be 60% in the combination group and 40% in the monotherapy group. These assumptions were based on the data obtained in the SONIC study.[1]

Analysis of the primary outcome measure included all patients randomly assigned to the monotherapy or combination groups, according to the intention-to-treat principle, with non-responder imputation of the data missing because of withdrawal, dropout or any other reasons. The same imputation method was used for the clinical remission at other time points [as secondary endpoints]. The intention-to-treat principle was also applied to the rates of clinical remission and clinical improvement at each time point. For the primary endpoint, Fisher's exact test was used for testing independency between the groups, and an odds ratio [OR] and its confidence interval [CI] were calculated using logistic regression model. Multiple logistic regression analysis was performed to estimate the OR adjusted for potential confounders and to search for factors independently associated with the linical remission at Week 26. Covariates included in the model were age, sex, body weight, the duration of the disease, disease location, previous surgery, presence of internal fistula, presence of anal fistula, smoking status and medication at entry [elemental diet, 5-aminosalicylic acid and steroids].

For SES-CD and other outcomes, prespecified per-protocol analyses were used. The rates of endoscopic response and positive AAA were compared between the two groups, using a chi-square test. Trough levels of ADA were compared, using an unpaired *t* test.

All patients who received at least one dose of the study drugs were included in the safety analysis. Safety comparisons were carried out between the monotherapy and combination groups using Fisher's exact probability test.

3. Results

3.1. Patients

During the predetermined period of recruitment from June 1, 2011 until June 31, 2014, 85 patients were randomly assigned to the combination group and 92 patients to the monotherapy group [Figure 1]. One of the patients was excluded from the study because the patient

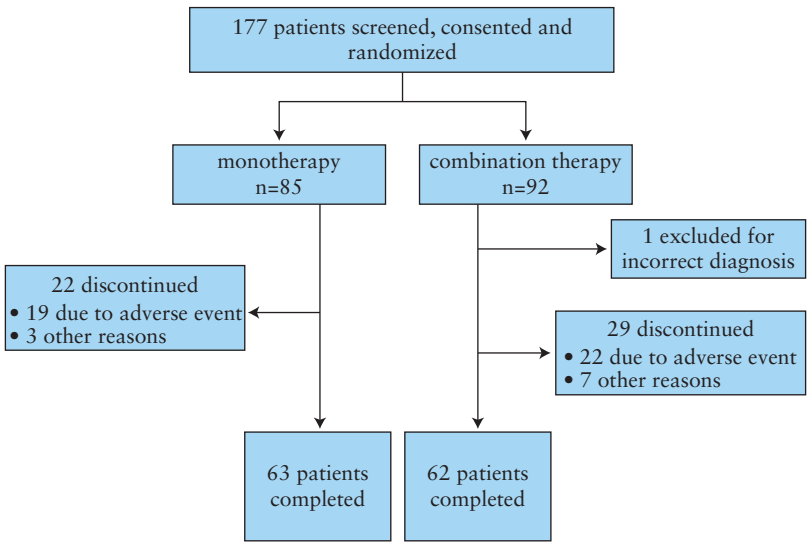

Figure 1. Enrolment and clinical course of the study population.

was diagnosed as having intestinal tuberculosis after the enrolment to the study; 63 patients in the monotherapy group and 62 patients in the combination group completed the study through Week 52. In the monotherapy group, 19 patients discontinued the study owing to adverse events, and three patients discontinued the study owing to other reasons [CDAI not available for two patients and consent withdrawal for one patient]. In the combination group, 22 patients discontinued the study due to adverse events and seven patients due to other reasons [dose escalation of AZA after 4 weeks for two patients, consent withdrawal for three patients and being lost to follow-up for two patients]. Table 1 summarizes and compares the demographic data and baseline clinical characteristics of the study population between the monotherapy group and combination groups.

3.2. Primary endpoint

The intention-to-treat analysis with non-responder imputation revealed that 61 of the 85 patients [71.8%] in the monotherapy group and 62 of the 91 patients [68.1%] in the combination group were in clinical remission at Week 26 (p = 0.63, odds ratio [OR]: 0.84, 95% confidence interval [CI]: 0.44–1.61] [Figure 2A]. The per-protocol analysis excluding 14 patients who discontinued the study due to side effects of the medications by Week 26, demonstrated a slightly higher remission rate in the combination group [62 of 78 patients, 79.5%] than in the monotherapy group [61 of 84 patients, 72.6%] [Figure 2B]. However, the difference did not reach statistical significance [p = 0.38, OR: 1.46, 95%CI: 0.71–3.03]. There was not any patient under steroid therapy in either group at Week 26.

A multiple logistic analysis was undertaken to estimate the ORs adjusted for potential confounders and to identify factors associated with clinical remission at Week 26. The adjusted OR was consistent with that obtained in the primary analysis [p = 0.48, OR: 0.77, 95% CI: 0.38–1.59]. Although there were trends for the clinical remission at Week 26 to be a negatively associated with the disease duration of 2 years or longer [vs less than 2 years] [p = 0.085, OR: 0.47, 95% CI: 0.20–1.11] and positively associated with the previous history of intestinal surgery [p = 0.088, OR: 2.88, 95% CI: 0.86–9.69], other variables including the AZA treatment were not associated with the clinical remission.

Table 1. Demographic and clinical characteristics of the patients.

	Monotherapy group [n = 85]	Combination group [n = 91]
Demographics		
Women	26 [31%]	24 [26%]
Age [years]	29 ± 12	32 ± 12
Duration of CD [years]	2.8 ± 5.9	3.2 ± 5.2
Disease location		
Ileitis [L1]	15 [15%]	19 [20%]
Ileocolitis [L2]	56 [67%]	58 [65%]
Colitis [L3]	14 [18%]	14 [15%]
Disease phenotype		
Inflammatory [B1]	36 [42%]	36 [40%]
Stricturing [B2]	28 [33%]	33 [36%]
Penetrating [B3]	21 [25%]	22 [24%]
Previous surgical resections		
0	76 [89%]	74 [81%]
1 or more	9 [11%]	17 [19%]
Current smoking	12 [14%]	14 [15%]
Medication at entry		
Elemental diet	38 [45%]	50 [55%]
5-ASA	64 [75%]	59 [65%]
Steroid use	13 [15%]	5 [6%]
C-reactive protein [mg/l]	26 ± 28	26 ± 25
CDAI	276 ± 62	265 ± 43
SES-CD	16 ± 8	15 ± 8

Data are n [%] or mean ± standard deviation.

5-ASA, 5-aminosalicylic acid; CD, Crohn's disease, CDAI, Crohn's Disease Activity Index; SES-CD, Simple Endoscopic Score for Crohn's disease.

3.3. Secondary endpoints

Figure 3 indicates the comparison data for the clinical remission rate [A] and clinical response rate [B] at Weeks 2, 4 12, 26 and 52. As shown in Figure 3A, the clinical remission rates were not different between the groups at Weeks 2, 12, 26 and 52. At Week 4, however, the rate was significantly higher in the monotherapy group [66 of 85 patients, 77.6%] than in the combination group [57 of 91 patients, 62.6%, p = 0.034]. The clinical response rates were not statistically different between the two groups at any time point.

The data for SES-CD were available for 175 patients at entry, for 115 patients at Week 26 and for 102 patients at Week 52. The mean [95% CI] SES-CD values at entry were 15.7 [14.0–17.3] in the monotherapy group [n = 90] and 14.7 [13.1–16.3] in the combination group [n = 85]. At Week 26, SES-CDs decreased to 7.8 [6.3–9.3] in the monotherapy group [n = 58] and to 5.7 [4.1–7.2] in the combination group [n = 57]. The SES-CD values at Week 52 were 7.3 [5.7–8.9] in the monotherapy group [n = 53] and 5.5 [3.8–7.2] in the combination group [n = 49]. As shown in Figure 4, the rate of endoscopic improvement at Week 26 was significantly higher in the combination group [84.2%] than in the monotherapy group [63.8%, p = 0.019]. However, the rates at Week 52 were not significantly different between the two groups [79.6% vs 69.8%, p = 0.36].

3.4. Safety profile

During the course through Week 52, adverse events leading to discontinuance of the study occurred in 19 patients [22.3%] in the monotherapy group and in 22 patients [24.2%] in the combination group [Table 2]. The incidences of the adverse events and their timings were not different between the two groups. Eighteen patients [21.2%] in the monotherapy group and seven patients [7.7%] in the combination group discontinued the study due to worsening of CD [p < 0.001]. In contrast, 15 patients [16.5%] in the combination group but only one patient [1.2%] in the monotherapy group discontinued the study because of side effects of the medications [p < 0.001].

3.5. 6-TGN, antibodies to adalimumab and adalimumab levels

6-TGN in RBCs was measured in 71 patients from the combination group at Week 12. The patients were administered AZA at doses ranging from 25 to 100 mg per day with a mean ± standard deviation [SD] of 0.86 ± 0.35 mg/kg. The 6-TGN levels ranged from 50 to 1510 pmol/8 × 10^8 RBCs, with the median [interquartile ranges] of 257 [162–426] pmol/8 × 10^8 RBCs. Per-protocol analysis revealed that 27 of 31 patients with 6-TGN < 250 pmol/8 × 10^8 RBCs [87.1%] and 33 of 34 patients with the level ≥ 250 pmol/8 × 10^8 RBCs [97.1%] were in remission at Week 26 [p = 0.13]. Similarly, 30 patients in the former group [96.8%] and 33 patients in the latter group [97.1%] showed clinical response at Week 26 [p = 0.43].

AAA and ADA trough levels were measured in 76 patients from the monotherapy group and in 75 patients from the combination group. AAA was positive in 10 patients [13.2%] from the monotherapy group

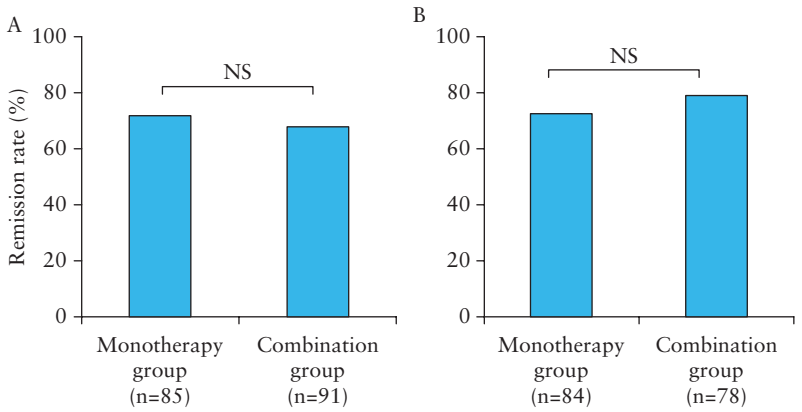

Figure 2. Comparison of clinical remission rates at Week 26. Intention-to-treat analysis with non-responder imputation [A], and per-protocol analysis, excluding patients, who discontinued the study because of side effects [B], demonstrate that the clinical remission rate did not differ between the monotherapy and combination groups.

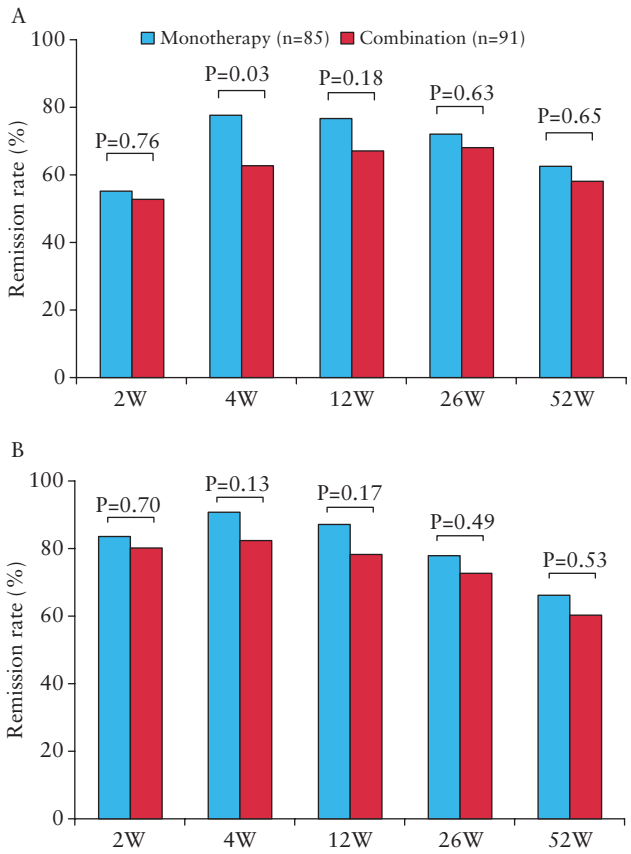

Figure 3. Rates of clinical remission [A] and clinical response [B].

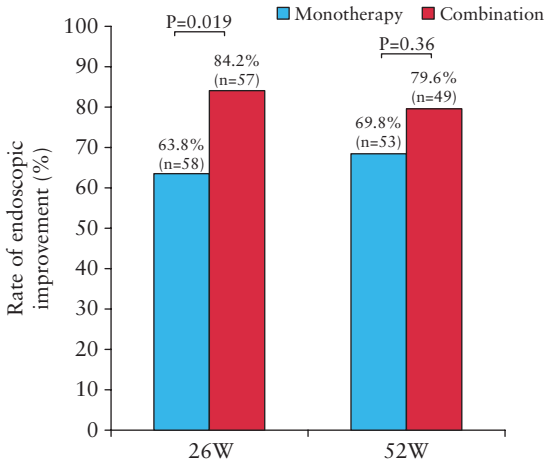

Figure 4. Comparison of the rates of endoscopic improvement at Weeks 26 and 52.

and in three patients [4.0%] from the combination group [p = 0.078]. The ADA trough level was 6.5 ± 3.9 µg/ml in the monotherapy group and 7.6 ± 3.6µg/ml in the combination group [p = 0.084]. Although not statistically significant, there were trends towards a higher ADA trough level and a lower positive rate of AAA in the combination group compared with those in the monotherapy group.

4. Discussion

The results of our randomized, open-labelled, prospective study indicated that the monotherapy with the use of ADA and the combination therapy with ADA and AZA were equally efficacious for intermediate-term maintenance of clinical remission in patients with CD, but the latter resulted in a better colonoscopic improvement at Week 26. It was also demonstrated that side effects of the study medications occurred more frequently in the combination group than in the monotherapy group.

The efficacy of the ADA and AZA combination still remains controversial. A stratified analysis of the data from a large clinical trial of ADA in patients with CD [CHARM trial] has shown that the induction and maintenance of remission were unrelated to the use of AZA.[2] In a retrospective analysis of 207 patients, Reenaers *et al.*[5] found no significant difference in the induction of remission between patients on a combination and those on ADA alone. In contrast, Peters *et al.*,[7] who analysed a cohort of 438 patients treated with ADA, found that the combined use of thiopurines contributed to a higher rate of continuance of ADA. In those analyses, however, there was heterogeneity in the timing of thiopurines, that is 23% of the study population had been taking thiopurines before the use of ADA. In a Hungarian cohort study, Kiss *et al.*[8] have shown that the concomitant use of AZA was an independent predictive factor for clinical remission at Week 52 after starting ADA. Based on a meta-analysis of these published data, Kopylov *et al.*[17] concluded that a combination of ADA and AZA was not superior to a monotherapy with ADA in terms of maintenance of remission at 1 year.

Table 2. Adverse events.

	Monotherapy group [*n* = 85]	Combination group [*n* = 91]	p-Value
Adverse event leading to discontinuation of study drug			
No. of patients [%]	19 [22.3%]	22 [24.2%]	0.94
Time duration from entry [days]	173 ± 97	100 ± 100	
Worsening of Crohn's disease			
No. of patients [%]	18 [21.2%]	7 [7.7%]	< 0.001
Time duration from entry [days]	178 ± 96	162 ± 112	
Side effect presumably associated with study drug			
No. of patients	1 [1.2%]	15 [16.5%]	< 0.001
Time duration from entry [days]	70	61 ± 71	
Aortitis	1 [1%]		
Leukocytepenia		4 [4%]	
Alopecia		3 [3%]	
Liver damage		3 [3%]	
Nausea		2 [2%]	
Fever		2 [2%]	
Appendicitis		1 [1%]	
Hyperamylasaemia		1 [1%]	
Lymphadenopathy		1 [1%]	
Other adverse event			
No. of patients	2 [2%]	6 [7%]	
Time duration from entry [days]	70 ± 87	136 ± 139	
Leukocytepenia		3 [3%]	
Nausea	1 [1%]	3 [3%]	
Skin eruption		1 [1%]	
Oedema		1 [1%]	
Liver damage	1 [1%]		

Our present study was the first prospective trial to examine the efficacy of the combination of ADA and AZA for patients with CD. Since all study subjects were naïve to both medications, and only 18 of the 176 subjects were receiving steroids, most of the subjects were regarded as being under a top-down strategy with the use of ADA. This seems to explain the high remission and response rates at Week 26 in both monotherapy and combination groups compared with those observed in a double-blind controlled trial of Japanese patients with CD.[3] Our results, together with those of the above-mentioned retrospective and cohort studies, suggest that AZA does not contribute to the enhancement of the efficacy of ADA treatment, unlike its contribution to the additional efficacy of IFX treatment.

Our results should be interpreted with caution with respect to the adverse events. Because 15 of the 91 patients in the combination group discontinued the study owing to side effects of the medications, the study seems to have been underpowered by the non-responder imputation. This seems to be especially the case for the significant difference in the remission rates at Week 4, because the side effects occurred predominantly until Week 4. However, it should also be noted that the discontinuance rate of AZA because of side effects in the present study [16.5%] was similar to the prevalence of intolerance to AZA in Japanese patients with ulcerative colitis.[18] It thus seems likely that the high discontinuance rate in the combination group observed in this study is not exceptional for the combination therapy with ADA and AZA in a clinical setting. Because the remission rate at Week 26 in the patients without serious side effects was slightly, but insignificantly, higher in the combination group [79.5%] than in the monotherapy group [72.6%] and discontinuation of the study due to active CD was more frequent in the latter than in the former, the simultaneous use of AZA might have had marginal efficacy for the maintenance of clinical remission in our study population.

There may be an argument that the doses of AZA applied to the combination group [25-100 mg/day] were lower that those used in the West. Although we did not measure 6-TGN at Week 26 and in fact 6-TGN level at Week 12 ranged widely, the dose of AZA given to our subjects remained unchanged through Week 52. Thus, the 6-TGN levels at Week 12 with the mean value within the appropriate range for therapeutic efficacy of thiopurines, and the incidence of adverse events in the combination group, suggest that the pharmacokinetics of AZA in our subjects was similar to that in the Western population. Accordingly, the insignificant difference in the clinical efficacy between the monotherapy and the combination groups does not seem to be specific to the Asian population, but rather it can be interpreted as a universal phenomenon. Furthermore, it seems possible that the marginal efficacy of thiopurines for patients receiving ADA may not be closely associated with 6-TGN level, since the remission and response rates at Week 26 were high and they were so regardless of 6TGN level at Week 12 among our patients in the combination group, who were able to complete the protocol through Week 26.

Mucosal healing has become another goal for the management of CD.[19] In the post hoc analysis of a prospective study that evaluated a combination of IFX and AZA, the combination resulted in a significantly higher rate of mucosal healing at Week 26 than the AZA monotherapy.[20] However, the additional efficacy of AZA for patients receiving ADA therapy remains obscure. Based on the results of our present investigation, it is presumed that the combination of ADA and AZA is superior to ADA monotherapy in obtaining mucosal healing at Week 26. Our result is consistent with the recent observation by Nuti *et al.*[21] who retrospectively showed the superiority of a combination of anti-TNF-α and AZA, compared with anti-TNF-α monotherapy, for mucosal healing in paediatric CD patients naïve to both medications. Since the endoscopic improvement at Week 52 did not differ between the monotherapy and combination groups, AZA may be associated with rapid mucosal healing in CD. These observations strongly suggest that a combination of ADA and AZA may be a beneficial treatment for CD patients with prominent mucosal

damage, and, provisionally, for the prevention of intestinal complications. This potential, as well as the achievement of deep remission with clinical, endoscopic and histological improvements,[22] needs to be investigated for the use of AZA in CD patients treated with ADA.

In addition to the clinical efficacy, we found marginal effects of AZA on the trough level of ADA and titres of AAA, with the trends of higher trough levels and a lower positive rate of AAA in the combination group. These observations suggest that thiopurines are protective against immunogenicity of ADA. The marginal efficacy of ADA in our study population may be associated with the marginal effect of AZA on immunogenicity of ADA.

There are several limitations of this study. First, the open-label nature of this investigation seems to have contributed to the increased number of subjects who discontinued the study. This seems to be especially true for the combination arm, and actually a greater number of subjects in the arm failed to complete the study because of side effects. Alternatively, the increased number of subjects with worsening of CD may be attributed to the open-label system. Second, we were unable to enrol a predetermined number of subjects. This actually resulted in statistical underpowering of this investigation. In this study, however, the observed remission rates were much higher than the initial hypothesis for the sample estimation. With the modified alternative hypothesis based on the result, such as '80% vs 60%', statistical power is 83.1% even with the final sample size. Because the statistical power is higher than the initial setting, our study seems to have had enough power to detect a risk difference of at least 20% unless biased discontinuation between the groups occurred.

In conclusion, the results of our prospective, open-label study indicated that the simultaneous use of AZA did not enhance the clinical efficacy of ADA for patients with CD within 1 year. This observation was based on the fact that the additional effect of AZA could not overcome its side effects. Considering mucosal healing, however, AZA may be an option for patients with CD, who are tolerant to the medication.

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Conflict of Interest

The authors have the following financial conflicts of interest regarding this manuscript. TM: Eisai Corporation [EC], Abbvie GK [AGK], Mitsubishi Tanabe Pharma [MTP]. SM: EC, AGK, MTP, Jannsen Pharma. KW: EC, AGK, MTP. TH: EC, AGK, MTP, Ajinomoto Pharma [AP]. HN: EC, AGK. NY: EC, AGK, MTP. SK: AGK. ME: EC, AGK, MTP. MN: EC, AGK, MTP. ToM: EC, AGK, MTP. YNa: Astellas Pharma [AsP], Otsuka Pharmaceutical Corporation [OPC], Takeda Pharmaceutical Corporation [TPO], EC, MTP. TK: EC, AGK, MTP. YS: EC, AGK, MTP, Zeria Pharmaceutical Corporation [ZPO]. MW: EC, AGK, MTP, Kyorin Pharmaceutical Corporation, Diichi Sankyo Corporation, Ono Pharmaceutical Corporation, Gene Care Research Institute, AsP, Asahi Kasei Kuraray Corporation, Chugai Pharmaceutical Corporation, TPO, AP, OPC, Kyowa Hakko Kirin Corporation, Jimro Corporation, ZPO, UCB Japan Corporation, Dainippon Sumitomo Pharma, Toray Industries, Bristol-Meyers KK. TH: [EC, AGK, MTP.

Author Contributions

Author contributions to the manuscript are as follows. Study concept and design: TM, KW, TH, HN, TK, YS, MW, TH; acquisition of data: SM, NY, TI, SK, TN, ME, MN, ToM, YN; analysis and interpretation of data: TM, KW, HT, HN, MNo; drafting the manuscript: TM, KW, HT, HN, MNo; critical revision of the manuscript for important intellectual content: MNa, YS, MW, TH; statistical analysis: MNo; and study supervision: YS, MW, TH.

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