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Yoichi Ichikawa · Terunobu Saito · Hisashi Yamanaka Masashi Akizuki · Hirobumi Kondo · Shigeto Kobayashi Hisaji Oshima · Shinichi Kawai · Nobuaki Hama Hidehiro Yamada · Tsuneyo Mimori · Koichi Amano Yasushi Tanaka · Yasuo Matsuoka · Sumiki Yamamoto Tsukasa Matsubara · Norikazu Murata · Tomiaki Asai Yasuo Suzuki · MTX–BUC Combination Study Group, Japanese Ministry of Health, Labour and Welfare's "Research for Establishment of Therapeutic Guidelines in Early Rheumatoid Arthritis"

Therapeutic effects of the combination of methotrexate and bucillamine in early rheumatoid arthritis: a multicenter, double-blind, randomized controlled study

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Abstract Disease-modifying antirheumatic drug (DMARD) combination therapies are used widely, but there have been few reports clearly demonstrating that combination therapy is more effective than DMARD monotherapy. We conducted a multicenter, double-blind controlled trial in order to clarify that the combination of methotrexate and bucillamine is more effective than either alone. The subjects of this study were 71 patients with active rheumatoid arthritis within 2 years of onset. Dosages were 8 mg methotrexate with 5 mg folic acid per week (MTX group), 200 mg bucillamine per day (BUC group), or both MTX and BUC (combination group). Clinical effects and adverse reactions were observed for 96 weeks. The ACR 20 response rate was 79.2% in the combination group, significantly higher than the rates of 43.5% for the MTX group ($P = 0.008$) and 45.8% for the BUC group ($P = 0.0178$). The cumulative survival curve of maintaining the ACR 20 response was significantly higher in the combina-

Y. Ichikawa[1] (✉) · N. Hama · H. Yamada · Y. Suzuki
Department of Rheumatic, Collagen and Allergic Diseases, St. Marianna University School of Medicine, 2-16-1 Sugao, Miyamae-ku, Kawasaki 216-8511, Japan

T. Saito · H. Yamanaka
Institute of Rheumatology, Tokyo Women's Medical University, Tokyo, Japan

M. Akizuki
Department of Internal Medicine, Yokohama Municipal Citizens' Hospital, Yokohama, Japan

H. Kondo
Department of Internal Medicine, Kitazato University School of Medicine, Kitazato, Japan

S. Kobayashi
Department of Internal Medicine, Juntendo University School of Medicine, Tokyo, Japan

H. Oshima
Department of Internal Medicine, Fujita Health University, Nagoya, Japan

S. Kawai
Institute of Medical Science, St. Marianna University School of Medicine, Kawasaki, Japan

T. Mimori
Department of Internal Medicine, School of Medicine, Keio University, Tokyo, Japan

K. Amano
Second Department of Internal Medicine, Saitama Medical Center, Kawagoe, Japan

Y. Tanaka
Department of Internal Medicine, Konan Kakogawa Hospital, Kakogawa, Japan

Y. Matsuoka
Department of Internal Medicine, Kawasaki Municipal Hospital, Kawasaki, Japan

S. Yamamoto
The Centre for Rheumatic Diseases, Matsuyama Red Cross Hospital, Ehime, Japan

T. Matsubara
Department of Orthopedics, Matsubara Mayflower Hospital, Hyogo, Japan

N. Murata
Department of Rheumatology, National Hospital Organization, Osaka Minami Medical Center, Osaka, Japan

T. Asai
Department of Orthopedics, National Hospital Organization, Nagoya Medical Center, Nagoya, Japan

Present address:
[1]St. Joseph's Hospital, 28 Midorigaoka, Yokosuka 238-8502, Japan
Tel. +81-46-822-2134; Fax +81-46-822-3134
e-mail: j-incho@aqua.ocn.ne.jp

tion group than in the MTX and BUC groups ($P = 0.0123$ and $P = 0.0088$, respectively). The mean increase in the total Sharp score over 96 weeks was 12.6 ± 9.0 in the combination group, significantly lower ($P = 0.0468$) than the value of 28.0 $\pm$ 28.3 for the single DMARD (combined MTX and BUC) group. The incidence of adverse reactions did not differ significantly between the three groups. It was concluded that the combination therapy with MTX and BUC showed significantly higher clinical efficacy than either of the single DMARD therapies.

Key words Bucillamine · Combination therapy · Methotrexate · Rheumatoid arthritis

Introduction

Although many studies have investigated the usefulness of combinations of disease-modifying antirheumatic drugs (DMARDs), very few studies have shown that a combination of two conventional DMARDs is more effective than the individual drugs alone.[1] For example, two double-blind controlled studies performed over 52 weeks showed no statistically significant increase in the disease amelioration rate with a combination of methotrexate (MTX) and sulfasalazine (SSZ).[2,3] Other combinations of DMARDs have also been investigated in double-blind controlled trials, but none of the combination therapies have been shown to be more effective than monotherapy.[4–7]

It was reported that the addition of cyclosporine was effective in subjects with inadequate control of rheumatoid activity by MTX.[8] As there was no comparison with treatment by cyclosporine alone, it was unclear, however, whether the effect of the combination therapy was the effect of the combination of MTX and cyclosporine, or the effect of cyclosporine itself. Several studies have also shown that the addition of a second conventional DMARD is effective in subjects with inadequate response to MTX,[9–11] but these are not thought to show exactly that combination therapies are more useful than monotherapy, as no comparisons were made of the effects of the combination and the additional DMARD alone. On the other hand, in comparison with MTX treatment alone, the combination of SSZ and hydroxychloroquine (HCQ), and the combination of these three drugs, O'Dell et al. found that simultaneous administration of the three DMARDs is the most effective.[12] Their study confirmed an additive or synergistic effect for this specific combination of DMARDs. There has, however, been no study showing by double-blind controlled trial that combination therapy with two conventional DMARDs is more effective than either DMARD administered as monotherapy.

In the present study, efficacy and adverse reactions were compared between MTX monotherapy, bucillamine (BUC) monotherapy, and the combination of both drugs, using a double-blind controlled trial, with the combination demonstrated to be the most effective. This is the first report showing that combination therapy with two conventional DMARDs is more effective than either DMARD administered as monotherapy.

Subjects and methods

The subjects were 71 Japanese patients aged from 20 to 70 years, within 2 years of the onset of continuous arthritic pain, who met the American College of Rheumatology (ACR) classification criteria for rheumatoid arthritis as revised in 1987. The subjects had 6 or more painful joints out of 48 and 3 or more swollen joints out of 46, and showed at least a 30 mm/h erythrocyte sedimentation rate (ESR), or 1.0 mg/dl or more of C-reactive protein (CRP). They had never received MTX or BUC treatment, or corticosteroids at dosages of 7.5 mg/day or more of prednisolone equivalent.

Twenty-three medical institutions participated in this study, and subjects entered the trial between February 1999 and March 2000. Subjects were divided into three groups, respectively administered MTX, BUC, or the combination of both drugs. After confirming a subject's eligibility for the study by telephone, one of the test drugs was assigned randomly according to the usage number for the test drug distributed beforehand to each institute. A placebo indistinguishable in appearance from the drug was prepared for MTX and BUC for a double dummy medication technique. Dosages were as follows: MTX 4 mg/week and BUC 100 mg/day for the first 4 weeks, then 6 mg/week and 200 mg/day respectively for the following 8 weeks, and 8 mg/week and 200 mg/day respectively from week 13 to week 96, the end of the study period. Folic acid 5 mg was administered 48 h after MTX or its placebo. The trial was discontinued for subjects who had not reached an ACR 20 response by 24 weeks, or who did not meet the ACR 20 response criteria thereafter. After withdrawal from the trial due to inadequate response, adverse reactions, etc., subsequent treatment was at the discretion of the treating physician. Planned observations, and laboratory and X-ray investigations were continued as scheduled until the end of the study period for these subjects.

The following observations and investigations were carried out at intervals of 4 weeks for each subject: physical examination, full blood count, hepatic transaminases, serum creatinine, blood urea nitrogen, and urinalysis. The following indices were determined to evaluate treatment effects just prior to study commencement and every 12 weeks thereafter for 98 weeks: number of painful joints, number of swollen joints, pain estimation by the subjects using the visual analogue scale (VAS), subject's global assessment of disease activity using the VAS, physician's overall assessment of disease activity using the VAS, the modified health-assessment questionnaire (mHAQ),[14] erythrocyte sedimentation rate (ESR) using the Westergren method, C-reactive protein (CRP), and rheumatoid factor (RF). Plain posterior-anterior radiographs of both hands were taken at the beginning of the study and 96 weeks later.

Table 1. Characteristics of patients with rheumatoid arthritis, according to study groups

	Methotrexate (n = 23)	Bucillamine (n = 24)	Combination (n = 24)
Female (%)	16 (69.6)	20 (83.3)	17 (70.8)
Age (years)	52.7 ± 9.3	52.5 ± 11.3	49.1 ± 12.9
Age range (years)	34–70	23–67	29–69
Height (cm)	157.9 ± 9.7	156.2 ± 7.2	158.8 ± 8.5
Weight (kg)	52.2 ± 9.0	53.3 ± 10.3	54.6 ± 8.9
Duration of illness (months)	8.2 ± 4.8	9.1 ± 5.3	10.6 ± 6.6
Steinbrocker stage (cases)			
Stage I	14	15	15
Stage II	9	9	9
Steinbrocker class (cases)			
Class I	4	2	3
Class II	16	18	19
Class III	3	4	2
Positive rheumatoid factor (%)	19 (82.6)	19 (79.2)	23 (95.8)
Corticosteroid therapy (%)	8 (34.8)	8 (33.3)	4 (16.7)
Dose (PSL eq) mg/day	4.6 ± 0.74	4.3 ± 1.4	4.8 ± 3.1
Previous DMARD (%)	4 (17.4)	9 (37.5)	4 (16.7)
No. of swollen joints	9.3 ± 5.9	11.5 ± 6.6	8.9 ± 4.3
No. of tender joints	15.9 ± 10.6	13.7 ± 8.3	13.6 ± 7.0
mHAQ	6.65 ± 3.72	5.79 ± 3.37	6.52 ± 3.70
Pain	67.0 ± 26.4	66.1 ± 26.3	61.5 ± 22.3
Global assessment[a]			
By patient	68.0 ± 21.8	66.7 ± 29.2	62.2 ± 22.0
By physician	69.8 ± 13.0	69.8 ± 22.8	65.6 ± 19.5
ESR (mm/h)	76.8 ± 39.3	71.7 ± 28.6	68.5 ± 33.7
CRP (mg/dl)	4.99 ± 4.31	4.13 ± 3.56	4.29 ± 4.53

PSL eq, prednisolone equivalent; DMARD, disease-modifying antirheumatic drug; mHAQ, Modified Health-Assessment Questionnaire; ESR, erythrocyte sedimentation rate; CRP, C-reactive protein

[a] Global assessment of disease activity

HLA-DRB1 genotype was determined at the beginning of the study.

Therapeutic efficacy was clinically evaluated based on the ACR 20 and 50 responses.[15] The progress of joint destruction was evaluated using the difference in the total Sharp score at the commencement of the study and 96 weeks later. The total Sharp score, the sum of the erosion score, and the joint space narrowing score of the plain radiographs of the hands was the mean of the scores determined individually by three rheumatologists (Y.I., N.H., H.Y.) using a modified Sharp method.[16]

Fisher's exact test was used to test significance in contingency table analyses. The Kaplan–Meier cumulative survival curve maintaining ACR 20 response was also compared between the three treatment groups, whereas the statistical significance was determined by log-rank test.

Results

Seventy-one subjects were included in this study, and data were analyzed on an intention-to-treat basis. Table 1 shows the background factors of the 23 subjects in the MTX group, 24 subjects in the BUC group, and 24 subjects in the combination group. The mean disease duration was 8.2, 9.1, and 10.6 months, respectively. The majority of subjects were in Stage I of Steinbrocker's staging, and nearly 40% were in Stage II. Many subjects were in functional status Class 2.

Rheumatoid factor was positive in 82.6%, 79.2%, and 95.8% of subjects, respectively. The mean swollen joint counts were 9.3, 11.5, and 8.9, and painful joint counts 15.9, 13.7, and 13.6, respectively. The mean ESR for each group ranged from 68.5 to 76.8 mm/h, whereas the mean CRP ranged from 4.13 to 4.99 mg/dl. There were no statistically significant differences between the three groups for any of these indices.

Table 2 shows the ACR 20 and 50 response rates of the three treatment groups at the final examination for each subject. The ACR 20 response rate in the combination group was 79.2%, significantly higher than the rate of 43.5% for the MTX group (P = 0.008) and 45.8% of the BUC group (P = 0.0173). The ACR 50 response rate was 58.3% in the combination group, 34.8% in the MTX group, and 37.5% in the BUC group, the differences being not statistically significant. The study was interrupted due to insufficient response for two subjects (8.3%) in the combination group, significantly fewer than the 10 subjects (43.5%) in the MTX group and 10 subjects (41.7%) in the BUC group (P = 0.0171 and P = 0.0355, respectively).

Figure 1 shows the cumulative survival curve of the subjects maintaining the ACR 20 response rate in each group by the Kaplan–Meier method. The cumulative survival rate

Table 2. ACR 20 and ACR 50 responses and discontinuation due to lack of efficacy observed during treatment in three treatment groups

	Methotrexate	Bucillamine	Combination
No. of cases	23	24	24
ACR 20 response (%)	10 (43.5)††	11 (45.8)†	19 (79.2)
ACR 50 response (%)	8 (34.8)	9 (37.5)	14 (58.3)
Discontinued due to lack of efficacy (%)	10 (43.5)**	10 (41.7%)*	2 (8.3%)

† $P = 0.0355$, †† $P = 0.0171$, * $P = 0.0173$, ** $P = 0.008$ vs combination

Table 3. Changes in total Sharp score over a 96-week period in patients treated for at least 24 weeks

	Methotrexate ($n = 15$)	Bucillamine ($n = 16$)	Combination ($n = 15$)
Total Sharp score at entry	24.3 ± 20.3	19.4 ± 14.4	14.4 ± 10.4
Changes in total Sharp score at 96 weeks	27.4 ± 31.2*	28.5 ± 26.2**	12.6 ± 9.0

* $P = 0.088$, ** $P = 0.034$ vs combination

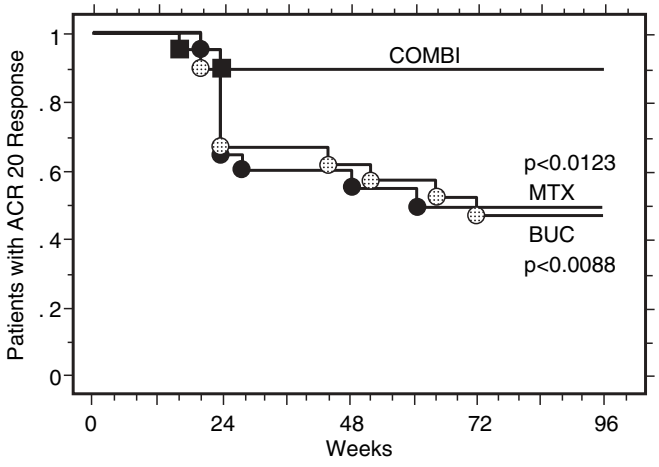

Fig. 1. Kaplan–Meier cumulative survival curve of the subjects maintaining the ACR 20 response rate. *Black circles*, methotrexate (*MTX*) group; *gray circles*, bucillamine (BUC) group; *squares*, combination (*COMBI*) group. The cumulative survival rate of the combination group was significantly higher than that of the MTX and BUC groups by log-rank test analysis ($P = 0.0123$ and $P = 0.0088$, respectively)

of the combination group was significantly higher than that in the MTX and BUC groups by log-rank test analysis ($P = 0.0123$ and $P = 0.0088$, respectively).

Table 3 shows the change in the total Sharp score over 96 weeks for subjects who continued the test drugs for more than 24 weeks. The increase in the total Sharp score during 96 weeks was 12.6 ± 9.0 in the combination group, lower than those of 27.4 ± 31.2 and 28.5 ± 26.2 in the MTX and BUC groups, respectively ($P = 0.088$ and $P = 0.034$), although there was no significant difference between the scores for each group at the beginning of the study. The increase in the total Sharp score in the combination group was also significantly lower than the value of 28.0 ± 28.3 for the single DMARD (combined MTX and BUC) group ($P = 0.0468$).

Treatment was discontinued due to adverse reactions in two subjects (8.7%) in the MTX group, six subjects (25.0%) in the BUC group, and seven subjects (29.2%) in the combination group, with no statistically significant differences between groups. The adverse reactions observed were as follows: one case each of leukopenia and proteinuria in the MTX group; three cases of rash, two of elevated transaminases, and one of proteinuria in the BUC group; and three cases of proteinuria and one each of elevated transaminases, rash, taste dyscrasia, and urinary tract infection in the combination group.

Discussion

Bucillamine is a sulfhydryl (SH) compound with a similar structure to D-penicillamine, but has two SH groups per molecule while D-penicillamine has one.[17] Both drugs suppress the proliferation and interleukin-2 production of T cells, although BUC is characteristically able to act without cupric ion,[18] and is currently widely used as an antrheumatic drug in Japan.

O'Dell et al. showed that simultaneous administration of MTX, SSZ, and HCQ is significantly more effective than MTX monotherapy or combination therapy with SSZ and HCQ.[12] They demonstrated an additive therapeutic effect for this combination of DMARDs. In addition, it has been reported that some combinations of biologic agents and a conventional DMARD, especially MTX, are more effective than treatment with an individual drug alone.[1,19]

On the other hand, there have been many reports of double-blind controlled studies investigating the usefulness of a combination of two conventional DMARDs, but none of these have shown statistically significant superiority for the combination compared to monotherapy.[2–7] In the present study, however, therapeutic efficacy was clearly shown to be higher in the combination group than in the

MTX or BUC groups, whereas the incidence of adverse reactions causing interruption of the study was not significantly different between the three groups.

The MTX dosage in this study was 8 mg/week, less than the dosages used in earlier studies.[12,20,21] The mean body weight of subjects in this study was, however, relatively light at 51.4 kg. The ACR 20 response rate at this dosage was 43.5%, consistent with reports of studies using MTX with[20] and without[21] folic acid. The ACR 20 response rate in the BUC group was 45.8%, similar to that in the MTX group, and 79.2% in the combination group, clearly higher than the other groups. The ACR 20 response rate in the combination group in this study was consistent with the 78% reported for the three DMARD combination therapy by O'Dell et al.[21]

Progress of bone destruction over the 96-week study period, as determined by Sharp's method modified by van der Heijde, was analyzed on an ITT (intention-to-treat) basis, giving a result of 31.7 ± 33.4 in the MTX group, 27.8 ± 31.0 in the BUC group, and 15.8 ± 15.8 in the combination group, with no statistically significant differences found between groups. Since the final dosage was reached at 13 weeks, it was considered appropriate to analyze all subjects administered test drugs for at least 24 weeks in order to compare the increases in the total Sharp score over 96 weeks between the three groups. The progress of bone destruction in the combination group was significantly lower than that of the BUC group, and showed a similar trend in comparison with the MTX group (Table 3). In addition, the increase in the Sharp score in the combination group was significantly lower than that in the combined MTX and BUC groups, i.e., the DMARD monotherapy group. These results suggest that combination therapy is superior to DMARD monotherapy not only in improvement of clinical symptoms, but also in suppression of bone destruction.

The present study demonstrated that the combination of MTX and BUC showed an additive clinical effect, using relatively small subject numbers, whereas earlier studies[2–7] had failed to show statistically significant superiority of combination therapies with various combinations of DMARDs. Bucillamine is known to suppress the function of T lymphocytes and B lymphocytes,[22] and inhibit the production of vascular endothelial growth factor (VEGF) in synovial cells.[23] On the other hand, MTX is known to suppress not only the function of lymphocytes[24,25] but also the function of neutrophils,[26] macrophages, and monocytes,[27] each of which has an important role in the pathogenesis of rheumatoid arthritis. The results of this study, showing increased therapeutic efficacy with combination therapy, suggest that these two DMARDs act on two or more different points of the pathogenetic process of rheumatoid arthritis.

Names of collaborating members and institutions: the MTX–BUC Combination Study Group, the Japanese Ministry of Health, Labour and Welfare's "Research for Establishment of Therapeutic Guidelines in Early Rheumatoid Arthritis"

Yoichi Ichikawa, Nobuaki Hama, Hidehiro Yamada, Yasuo Suzuki, Shouichi Ozaki (Department of Rheumatic, Collagen and Allergic Diseases, St. Marianna University School of Medicine), Terunobu Saito, Hisasi Yamanaka (Institute of Rheumatology, Tokyo Women's Medical University), Masashi Akizuki, Yutaka Okano (Department of Internal Medicine, Yokohama Municipal Citizens' Hospital), Hirobumi Kondo, Touru Akaboshi (Department of Internal Medicine, Kitasato University School of Medicine), Hisaji Oshima (Department of Internal Medicine, Fujita Health University School of Medicine), Shigeto Kobayashi (Department of Internal Medicine, Juntendo University School of Medicine), Takao Fujii, Michito Hirakata, Tuneyo Mimori (Department of Internal Medicine, School of Medicine, Keio University), Koichi Amano (Second Department of Internal Medicine, Saitama Medical Center), Yasushi Tanaka (Department of Internal Medicine, Konan Kakogawa Hospital), Takahiro Suzuki, Yasuo Matsuoka (Department of Internal Medicine, Kawasaki Municipal Hospital), Nobuo Takubo, Sumiki Yamamoto (The Centre for Rheumatic Diseases, Matsuyama Red Cross Hospital), Tsukasa Matsubara (Department of Orthopedics, Matsubara Mayflower Hospital), Norikazu Murata (Department of Rheumatology, National Hospital Organization, Osaka Minami Medical Center), Tomiaki Asai (Department of Orthopedics, National Hospital Organization, Nagoya Medical Center), Yasuyuki Miyata, Yukio Sato (Second Department of Internal Medicine, Fukushima Medical University), Norihiro Nishimoto (School of Health and Sport Science, Osaka University), Eizo Saito (Fourth Department of Internal Medicine, School of Medicine, Toho University), Shigemasa Sawada (Nerima-Hikarigaoka Hospital, School of Medicine, Nihon University), Akira Rikimaru (National Hospital Organization, Morioka Hospital), Hajime Yamagata (National Hospital Organization, Murayama Medical Center), Shigeto Toma (Department of Internal Medicine, National Hospital Organization, Sagamihara National Hospital), Katsumi Ito (Department of Rheumatism and Collagen Diseases, Metropolitan Bokuto Hospital), Shinichi Kawai (Institute of Medical Science, St. Marianna University), Katsumi Yoshida (Department of Preventive Medicine, St. Marianna University).

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