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Interventional Evaluation of Monoammonium Glycyrrhizinate-Glycine/DL-Methionine Combination Tablets in Mild Alopecia Areata

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Abstract

Objective: Although monoammonium glycyrrhizinate/glycine/DL-methionine (MG) combination tablets have been widely used widely for the treatment of alopecia areata (AA), there are few studies on efficacious combinations with MG. This study was conducted to determine the efficacy and safety of MG plus 5% carpronium chloride (CC).

Methods: In the present interventional study, MG tablets plus 5% carpronium chloride (CC) were compared with CC monotherapy in 31 patients with AA.

Results: There were no significant differences in efficacy between the two groups, and the AA area at 8 and 12 weeks was significantly reduced in both. The results of subanalysis stratified by the presence of allergic factors as determined by IgE level showed that there were also significant decreases in the areas of AA 8 and 12 weeks after the start of the combination therapy in patients with allergic factors ($p<0.05$). No serious adverse events were observed in either group.

Conclusion: It is suggested that combination therapy with MG and CC has better therapeutic effects than CC monotherapy, with a significant decrease in the area of AA from 4 weeks of treatment even in mild AA patients with allergic factors.

Keywords: Alopecia areata; Glycyrrhizinate-glycine/DL-methionine combination tablets; Carpronium chloride; Allergic factors; IgE

Introduction

Alopecia areata (AA) is the most frequent form of acquired alopecia. It was reported that it has a 0.1-2% prevalence rate in the USA, with a lifetime prevalence rate of 1.7%, and those figures are estimated to be comparable in Japan [1,2]. This condition can occur in all ethnic groups, genders, and age-groups. Recently, genes involved in the development of AA have been identified. A tendency for AA to occur in severe atopic dermatitis patients with filaggrin gene abnormalities has been reported, a family history is often seen, and therefore AA is considered to be a multifactorial genetic disease [3-5]. In addition, there is a high rate of concomitant atopic disease and autoimmune disease [6-8]. Even in the initial stage of AA, some studies found that the IgE level is elevated [9,10]. It is well known that glycyrrhizin has antiinflammatory effects, and thus the effects of monoammonium glycyrrhizinate/glycine/DL-methionine (MG) on AA may be related to immunological activity. The hair follicle has immunological privilege (IP) and is not susceptible to attack by normal immunocompetent cells. Theoretically, the autoimmune reaction to the hair follicle tissue due to the collapse of the IP is the main etiology of AA, which can be triggered by widespread infection, autoimmune disease, or psychological stress in those with a genetic predisposition to develop it [11,12].

Various treatments for AA have been proposed, but many cases relapse and/or prove refractory, and thus the establishment of appropriate treatment is necessary. The Japanese Dermatological Association Alopecia Areata Clinical Practice Guidelines were proposed in 2010 and recommend treatment according to the severity and stage of disease [13]. MG tablets have been used for the treatment of AA for more than 50 years, and those guidelines state that they can also be used in combined treatment. However, there are few detailed reports on agents that can be combined with MG tablets for the treatment of this condition. We therefore performed a preliminary investigation of the appropriate use of MG tablets for the treatment of AA and compared the efficacy and safety of combined treatment with MG tablets and carpronium chloride (CC; Furozin) solution and treatment with CC solution alone.

Patients and Methods

Outpatients with AA who met the following criteria were enrolled in this study: five or fewer separate areas of hair loss or a hair loss area of less than 25% of the scalp; and aged 20 years or older who gave written informed consent for study participation. Exclusion criteria were: severe dermatitis or eczema of the scalp; hair loss score of 3 or more in the pull test; received oral, inhalation, or local-injection

steroids or topical treatment of the scalp within 1 month prior to the study; the use of agents to promote hair growth or prevent hair loss within 1 month prior to the study; a history of allergy to glycyrrhizin or carpronium; a diagnosis of malignancy; complications from severe liver, kidney, or heart disease; pregnant, potentially pregnant, hoping to become pregnant, or lactating during the study period; and deemed ineligible by the attending physician for other reasons.

Eleven patients were assigned to the single-treatment group and 20 to the combined-treatment group using the minimization method to ensure a balance between the two factors of age and number of areas of hair loss. In the single-treatment group, the recommended dosage of CC was applied to the affected area twice daily (morning and evening). In the combined-treatment group, the recommended dosage of CC solution was applied to the affected area twice daily (morning and evening) and 3. It contains the components in one tablet.as monoammonium glycyrrhizinate 35 mg (glycyrrhizinate 25 mg), Glycine 25 mg, DL-methionine 25 mg. MG tablets were taken three times daily (after meals). During the study period, the patients did not receive any other drugs for the treatment of AA. In addition, pharmaceutical products likely to induce hirsutism side effects such as minoxidil, steroids, and cyclosporine; antiandrogenic agents such as finasteride; antihistamines, other drugs, and quasi-drugs; and cosmetics promoting hair growth or hair loss prevention were prohibited.

For all patients, date of birth, gender, number of areas of hair loss, size of areas of hair loss, results of the pull test, and second-degree family history of AA or atopic predisposition were recorded in addition to meeting the exclusion criteria and serum IgE levels. When more than one area of hair loss was present, the area of maximum hair loss was defined as the area for observation at the start of the study.

Item	Comparison with study start	Evaluation	Score
Number of areas of hair loss	Increased	Worsened	-1
	No change	No change	0
	Decreased	Improved	1
Size of area (area observed) of hair loss	Increased	Worsened	-1
	No change	No change	0
	Decreased	Improved	1
	Disappeared	Markedly improved	2
Degree of hair breakage and callous	Increased	Worsened	-1
	No change	No change	0
	Decreased	Improved	1
	Disappeared	Markedly improved	2
Terminal hair growth	No Changing	No change	0
	Growing at low density	Improved	1
	Growing at high density	Markedly improved	2

Table 1: Physicians' findings.

At the beginning of the study, and at 4, 8, and 12 weeks, the attending physician recorded the number of areas of hair loss, size of the areas, degree of hair breakage, and callous hair inside and outside the observed areas, in addition to the growth of terminal hair. At each evaluation visit, the attending physician compared the patients' conditions with that at the beginning of treatment and assigned scores (Table 1).

The areas observed were also photographed at each visit. The scores at 4, 8, and 12 weeks were summed, and the treatment effect was assessed as shown in Table 2. In addition, at 4, 8, and 12 weeks, the attending physician asked all patients about their impressions of the treatment effects compared with their conditions at the start of the study, and their responses were recorded in the same six categories (Table 3).

Evaluation	Criteria	Overview of effect
Worsened	Total score –1	Progressive hair loss
No change	Total score 0	No change
Slightly improved	Total score 1–2	Trend toward improvement
Moderately improved	Total score 3–4	Developing improvement
Markedly improved	Total score 5	Improvement
Not determined	Missing data	

Table 2: Treatment effect evaluation.

At the eligibility survey, beginning of the study, and at 4, 8, and 12 weeks, the attending physician interviewed the patients on their general health and recorded subjective symptoms and objective findings reported. Pulse rates and blood pressure were measured, and laboratory blood testing was performed at the beginning of the study and at 12 weeks.

Evaluation	Impression
Worsened	Progressive hair loss
No change	No change
Improved	Terminal hair growth
Markedly improved	Nearly recovered
Not determined	Missing data

Table 3: Patients' impression of treatment effects.

At 12 weeks, the attending physician made a comprehensive evaluation of the six categories of treatment effects, taking into account the patients' own impressions, photographic evidence, the occurrence of side effects, and other data including subjective symptoms, objective findings, blood pressure, pulse rate, and blood test results. This comprehensive evaluation was the primary endpoint of the study. The safety evaluation was based on the incidence of adverse events including abnormal changes in blood test results and side effects for which a causal relationship with the study drugs could not be ruled out. As the secondary endpoints, efficacy and safety were evaluated

separately. In addition, the reduction in areas of hair loss was evaluated by measuring each area using Image J 1.47v software and comparing them with the areas at the start of the study.

Statistical analysis

Statistical analyses of the comprehensive evaluation, patients' impressions, and treatment effects were performed using Fisher's exact test on the results aggregated for the frequency of the categorical variables in the contingency table. Changes in the area of hair loss and physicians' findings were compared in the rank-sum Wilcoxon test. In the safety evaluation, the χ^2 test was used to determine the incidence of adverse events and Student's t-test for blood test results, blood pressure, and pulse rate changes. The statistical analysis software SAS ver. 9.2 or later (SAS Institute, Cary, NC, USA) was used.

	Single treatment					**Combined treatment**				**p-value**	**Note**
		Average		SD			Average		SD	-5	
				%					%		
Age	11	43.9	±	11.2		20	42.7	±	13.2	0.7909	1
Gender											
Male		2		18.2			6		30	0.4634	2
Female		9		81.8			14		70		
Number of areas of hair loss											
Average	11	2.1	±	1.6		20	1.4	±	0.6	0.0681	1
1		7		63.6			14		70	0.039	3
2		0		0			5		25		
3		1		9.1			1		5		
4		2		18.2			0		0		
5		1		9.1			0		0		
Area of hair loss											
<25		11		100			20		100	-	2
≥ 25		0		0			0		0		
Pull test											
2		11		100			20		100	-	2
3		0		0			0		0		
Family history of alopecia areata											
Yes		2		18.2			4		20	0.9021	2
No		9		81.8			16		80		
Atopic predisposition											
Yes		4		36.4			4		20	0.3255	2
No		7		63.6			16		80		
Lactating											
No		9		81.8			14		70	-	4
Pregnant											
No		9		81.8			14		70	-	4
History of oral steroid treatment											
Yes		0		0			0		0	-	2

No		11		100			20		100		
Topical steroid treatment for alopecia areata											
Yes		0		0			1		5	0.3442	2
No		11		100			19		95		
Allergy to carpronium or glycyrrhizin											
Yes		0		0			0		0	-	2
No		11		100			20		100		
Dermatitis and eczema on scalp											
Yes		0		0			0		0	-	2
No		11		100			20		100		
Diagnosis of malignancy											
Yes		0		0			0		0	-	2
No		11		100			20		100		
Severe liver, kidney, or heart disease											
Yes		0		0			0		0	-	2
No		11		100			20		100		
Concomitant drug											
Yes		0		0			4		20	0.0505	2
No		11		100			16		80		
IgE RIST		117.4	±	156.4			218.2	±	391.6	0.4225	1

Notes

1. Comparison between groups using Student's t-test.
2. Comparison between groups using the χ^2 test.
3. Comparison between groups using Fisher's exact test.
4. Women only.
5. -, p-value not calculated.

Table 4: Patient characteristics.

Result

Patient characteristics

The characteristics of patients are shown in Table 4. The single-treatment group had more areas of hair loss than the combined-treatment group at the start of the study (p=0.0390). Four patients in the combined-treatment group had received drugs other than the study drugs (p=0.0505). No significant differences were found in other characteristics between the two groups.

Evaluation	Single treatment		Combined treatment		p-value
	n	%	n	%	
Markedly improved	4	36.4	4	21.1	0.7954
Moderately improved	3	27.3	8	42.1	
Slightly improved	4	36.4	5	26.3	

No change	0	0	1	5.3	
Worsened	0	0	1	5.3	
Not determined	0	0	0	0	
Total	11	100	19	100	-

Table 5: Comprehensive evaluation. Comparison between groups using Fisher's exact test.

Efficacy evaluation (primary endpoint)

The comprehensive evaluation results evaluated by the attending physicians (11 patients in the single-treatment group and 19 in the combined-treatment group after 2 dropped out of the study after 8 weeks) are shown in Table 5. No significant difference between the two groups was seen. Seven of 11 patients (63.6%) in the single-treatment group and 12 of 19 (63.2%) in the combined-treatment group showed moderate or greater improvement.

		4 weeks		8 weeks		12 weeks	
Single Treatment		n	%	n	%	n	%
	Yes	1	9.1	0	0	0	0
	No	10	90.9	11	100	11	100
Combined Treatment	Total	11	100	11	100	11	100
	Yes	3	15.8	1	5.9	0	0
	No	16	84.2	16	94.1	17	100
	Total	19	100	17	100	17	100
p-value		0.5939		0.3121		-	

Table 6: Adverse events during the study period [Comparison between groups using the χ^2 test].

Safety evaluation (primary endpoint)

The number and incidence rate of adverse events at 4, 8, and 12 weeks; the number and incidence rate of subjective symptoms and objective findings; and the blood test results at the beginning of the study and at 12 weeks are shown in Tables 6-9, respectively. Seven adverse events (1 in the single-treatment and 6 in the combined-treatment groups) occurred in 5 patients (1 in the single-treatment group and 4 in the combined-treatment groups), and a causal relationship with the study drugs could not be ruled out in 3 of those adverse events (all in the combined-treatment group). The 3 adverse events that were possibly associated with the study drugs were 2 episodes of headache and 1 of itching at the topical application site. The symptoms were mild, and all patients recovered.

		Start		4 weeks		8 weeks		12 weeks	
		n	%	n	%	n	%	n	%
Single treatment	Yes	0	0	0	0	0	0	0	0
	No	11	100	11	100	11	100	11	100
	Total	11	100	11	100	11	100	11	100
Combined Treatment	Yes	1	5	2	10.5	1	5.9	1	5.9
	No	19	95	17	89.5	16	94.1	16	94.1
	Total	20	100	19	100	17	100	17	100
p-value		0.3442		0.1671		0.3121		0.3121	

Table 7: Subjective symptoms reported [Comparison between groups using the χ^2 test].

Three patients dropped out of the study, 1 after 4 weeks and 2 after 8 weeks. The compliance of other patients was good, with no deviation in dosage and administration. Data on the patients who dropped out were aggregated and treated as missing values.

		Start		4 weeks		8 weeks		12 weeks	
Single		n	%	n	%	n	%	n	%
treatment	Yes	0	0	0	0	0	0	0	0
	No	11	100	11	100	11	100	11	100
	Total	11	100	11	100	11	100	11	100
Combined		n	%	n	%	n	%	n	%
treatment	Yes	1	5	1	5.3	1	5.9	0	0
	No	19	95	18	94.7	16	94.1	17	100
p-value		0.3442		0.334		0.3121			

Table 8: Objective findings [Comparison between groups using the χ^2 test].

Changes in findings, treatment evaluation, and patient impressions (secondary endpoints)

Changes in physicians' findings, evaluation of treatment, and patients' impressions at 4, 8, and 12 weeks compared with those at the start of study are shown in Tables 10-12, respectively. No significant differences between the two groups were seen throughout the study period.

Single treatment									
Item	**Start**				**12 weeks**				
	n	Ave.	±	SD	n	Ave.	±	SD	p-value1
SBP	11	115.5	±	27.1	11	112.3	±	18.6	0.508
DBP	11	74.3	±	16.3	11	72.6	±	12.5	0.6382
Puls	11	73.3	±	11	11	69.7	±	12.9	0.3365
BUN	11	14	±	4.9	11	13.3	±	2.8	0.5577
CRE	11	0.64	±	0.09	11	0.65	±	0.1	0.4579
AST	11	19	±	3.4	11	19.2	±	4.8	0.8591
ALT	11	16	±	4.4	11	17.2	±	4.1	0.4802
ALP	11	189	±	28.8	11	185.5	±	36.1	0.556
LDH	11	193.3	±	61.6	11	182.1	±	29.4	0.4305
γ-GTP	11	22	±	10	11	19.7	±	7.8	0.203
CRP	11	0.05	±	0.03	11	0.08	±	0.09	0.2745
WBC	11	6554.5	±	1465.9	11	5990.9	±	1363.4	0.1034
RBC	11	451.2	±	41.9	11	450.6	±	41.1	0.9334
Hgb	11	13.8	±	1.3	11	13.7	±	1.2	0.6116
PLT	11	26.9	±	6	11	25.5	±	6.2	0.242
	11	100	±	0	11	100	±	0	-
Eos	11	1	±	1	11	0.8	±	0.8	0.6761
Baso	11	2.1	±	1.8	11	3	±	2.6	0.4171

Neut-Stab	11	2	±	1	11	1.6	±	0.8	0.3705
Neut-Seg	11	58.9	±	11.1	11	50.8	±	6	0.0569
Ly	11	32	±	9.1	11	37.9	±	7	0.1246
Mono	11	4	±	2.3	11	5.8	±	3.7	0.1852

Table 9: (a) Showing single treatment blood test results.

Combined treatment													
Item	**Start**				**12 weeks**				**Abort**				
	n	Ave.	±	SD	n	Ave.	±	SD	n	Ave.	±	SD	p-value1
SBP	20	118.2	±	24.1	17	117.2	±	13.7	2	127	±	32.5	0.687
DBP	20	77.5	±	15.1	17	76.6	±	13.9	2	72.5	±	3.5	0.777
Puls	20	76.2	±	14.4	17	75.4	±	11.5	2	66	±	5.7	0.8455
BUN	20	14.5	±	3.1	17	13.6	±	3.1	2	10.5	±	3.5	0.1762
CRE	20	0.67	±	0.1	17	0.72	±	0.14	2	0.6	±	0.1	0.003
AST	20	22.8	±	9.2	17	21.9	±	11.8	2	19	±	1.4	0.1441
ALT	20	23.2	±	15	17	23.9	±	20.1	2	13	±	1.4	0.6551
ALP	20	188.6	±	38.6	17	195.2	±	37	2	191	±	67.9	0.3037
LDH	20	168.7	±	29.2	17	174.7	±	27.5	2	173.5	±	33.2	0.6236
γ-GTP	20	29	±	25.3	17	29.5	±	24.8	2	14.5	±	3.5	0.3906
CRP	20	0.07	±	0.08	17	0.08	±	0.09	2	0.04	±	0.01	0.4484
WBC	20	5315	±	1458.3	17	5705.9	±	2011.7	2	4050	±	1343.5	0.4471
RBC	20	450.2	±	51	17	454.9	±	57.5	2	428.5	±	26.2	0.8707
Hgb	20	13.5	±	1.5	17	13.6	±	1.6	2	13.4	±	1.8	0.7747
PLT	20	24.2	±	4.2	17	24.2	±	5	2	25.4	±	3	0.8832
	20	100	±	0	17	100	±	0	2	100	±	0	-
Eos	20	0.9	±	0.9	17	0.8	±	0.9	2	2	±	1.4	0.8899
Baso	20	2.7	±	2.2	17	4.9	±	4.3	2	5	±	7.1	0.0765
Neut	1	55.1	-	-		-	-			-	-		-
Neut-Stab	19	1.5	±	0.6	16	1.6	±	0.7	2	2	±	0	0.5805
Neut-Seg	19	56.5	±	7.8	16	53.8	±	8.1	2	55.5	±	12	0.3085
Ly	20	33.9	±	7.7	17	33.6	±	6.9	2	30.5	±	4.9	0.897
Mono	20	4.8	±	2.3	17	5	±	1.6	2	5	±	1.4	0.6925

1. Comparison between start and 12 weeks within each group using the paired t-test.
2. Comparison between groups using the paired t-test (start).
3. Comparison between groups using the paired t-test (12 weeks).

Table 9: (b) Showing combined treatment blood test results.

Between groups	**p-value2**	**p-value3**

0.7848	0.4283
0.5895	0.4446
0.5636	0.2387
0.753	0.7507
0.4678	0.1632
0.2019	0.4712
0.1326	0.2837
0.9763	0.497
0.1407	0.5051
0.3886	0.2207
0.5107	0.9164
0.0315	0.6841
0.9548	0.8339
0.5729	0.7551
0.1522	0.5415
-	-
0.7063	0.9869
0.4681	0.2024
-	-
0.0832	0.8063
0.4943	0.306
0.5516	0.119
0.378	0.4285

Table 9: Blood test results.

Changes in areas of hair loss (secondary endpoint)

Areas of hair loss at 4, 8, and 12 weeks in 11 patients in the single-treatment group and 17 in the combined-treatment group, excluding the patients who dropped out, were compared with the areas at the start of the study. No significant difference was seen between the two groups. Although no reduction in the size of areas of hair loss was observed in the single-treatment group at any time point, a reduction was observed in the combined-treatment group at 4 weeks and thereafter (Table 13 and Figure 1).

In patients with allergic factors, as indicated by "IgE ≥ 171" in Table 14, comprising 6 in the single-treatment group and 7 in the combined-treatment group, the areas of hair loss at each observation were compared with those at the start of the study. In the combined-treatment group, the areas of hair loss at 8 and 12 weeks were reduced significantly (both $p<0.05$, Table 14, Figure 1).

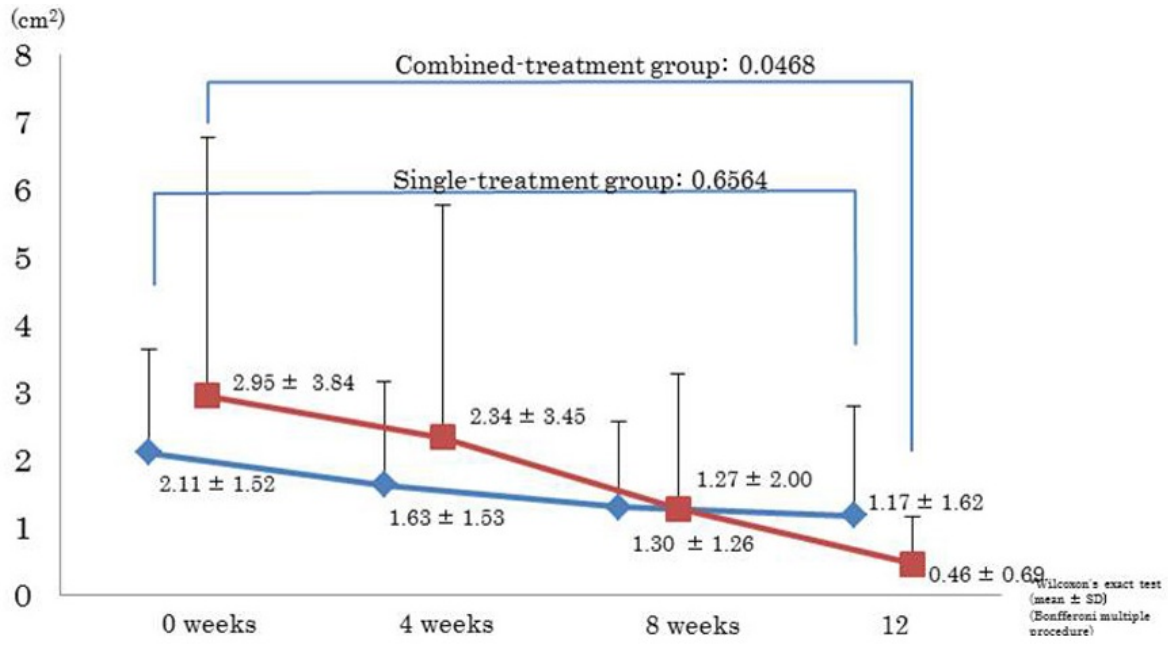

Figure 1: Change in size of areas of hair loss in patients with atopic predispositions in the combined-treatment and single-treatment groups at the start of the study and at 12 weeks.

Discussion

The Alopecia Areata Clinical Practice Guidelines were formulated based on the results of extensive clinical practice and other recommendations [11]. However, additional treatment options are required because the condition has a major effect on patients' quality of life, refractory and recurrent cases are common, and regeneration of

the hair may take a long time. MG tablets have been used widely for many years in treating AA and are recommended as combined therapy in the clinical practice guidelines. This study investigated combined treatment with MG tablets and CC solution. It was reported that this combination showed efficacy in 60% of AA patients with moderate or more severe disease [14]. However, no randomized, controlled study evaluating the efficacy of MG tablets has been conducted, as pointed out in the guidelines.

Start to 4 weeks											
	Single treatment					**Combined treatment**					**p-value**
Items	n	Ave.		SD	Median	n	Ave.		SD	Median	
Number of areas of hair loss	11	0	±	0	0	19	-0.1	±	0.2	0	0.4891
Size of area of hair loss	11	0.4	±	0.8	1	19	0.1	±	0.6	0	0.2413
Hair breakage and callous	11	0.5	±	0.7	0	19	0.2	±	0.4	0	0.3283
Terminal hair growth	11	0.8	±	0.6	1	19	0.7	±	0.7	1	0.7149
Total score	11	1.6	±	1.6	2	19	1	±	1.2	1	0.3149
Start to 8 weeks											
Number of areas of hair loss	11	0	±	0	0	17	0.1	±	0.4	0	0.6613
Size of area of hair loss	11	0.9	±	0.3	1	17	0.6	±	0.8	1	0.236
Hair breakage and callous	11	0.5	±	0.7	0	17	0.6	±	0.6	1	0.3565
Terminal hair growth	11	1.4	±	0.7	1	17	1.1	±	0.7	1	0.3655
Total score	11	2.7	±	1.1	3	17	2.4	±	1.9	2	0.5755
Start to 12 weeks											
Number of areas of hair loss	11	0.4	±	0.5	0	17	0.3	±	0.7	0	0.9155
Size of area of hair loss	11	1.3	±	0.6	1	17	0.9	±	1	1	0.4417
Hair breakage and callous	11	1.2	±	0.9	1	17	1.1	±	0.9	1	0.8603
Terminal hair growth	11	1.3	±	0.8	1	17	1.2	±	0.7	1	0.8371
Total score	11	4.1	±	2.6	3	17	3.6	±	2.6	3	0.7558

Table 10: Physicians' findings [Comparison between groups using the Wilcoxon rank-sum test].

Therefore, we compared the effectiveness of MG tablets alone with the combination of MG tablets and CC solution. No significant differences in efficacy, clinical findings, and patients' impressions of efficacy were observed between the two groups. A significant reduction in areas of hair loss was not observed in the single-treatment group but was observed in the combined treatment group at 4 weeks and thereafter. This result may have been due to the difference in the number of patients (11 versus 17) in the two groups. No serious adverse events occurred during the study, and the incidence of adverse events in the two groups did not differ significantly.

It has been reported that the IgE level as an allergic factor is elevated in the early stage of AA [15] .Therefore we conducted an analysis by allergic factors. The area of AA was significantly decreased in patients with allergic factors 8 and 12 weeks after treatment as compared with baseline, while a reduction in the AA area was observed in patients without allergic factors but was not significant.

4 weeks					
	Single treatment		**Combined treatment**		**p-value**
Evaluation	n	%	n	%	
Worsened	1	9.1	2	10.5	0.8064
No change	2	18.2	6	31.6	

Slightly improved	6	54.5	9	47.4	
Moderately improved	1	9.1	2	10.5	
Markedly improved	1	9.1	0	0	
Total	11	100	19	100	-
8 weeks					
Worsened	0	0	1	5.9	
No change	0	0	2	11.8	
Slightly improved	5	45.5	6	35.3	0.8539
Moderately improved	5	45.5	6	35.3	
Markedly improved	1	9.1	2	11.8	
Total	11	100	17	100	-
12 weeks					
Worsened	0	0	2	11.8	
No change	1	9.1	0	0	
Slightly improved	2	18.2	3	17.6	0.6644
Moderately improved	3	27.3	6	35.3	
Markedly improved	5	45.5	6	35.3	
Total	11	100	17	100	-

Table 11: Treatment evaluation [Comparison between groups using Fisher's exact test].

It was shown in vitro that glycyrrhizin acid, which is the active ingredient in MG tablets, is hydrolyzed by β-D-glucuronidase and metabolized to glycyrrhetinic acid [16,17]. Since glycyrrhetinic acid has an inhibitory effect on 11β-HSD2, the enzyme that metabolizes inactive cortisone to cortisol, it is possible that the antiinflammatory effects of cortisol in the body are indirectly affected by glycyrrhetinic acid, resulting in improved AA [18].

	Single treatment		**Combined treatment**		**p-value**
Evaluation	n	%	n	%	
4 weeks					
Worsened	0	0	2	10.5	
No change	6	54.5	6	31.6	
Improved	5	45.5	10	52.6	0.5851
Markedly improved	0	0	1	5.3	
Not determined	0	0	0	0	
Total	11	100	19	100	-
8 weeks					
Worsened	0	0	1	5.9	
No change	1	9.1	4	23.5	0.6832
Improved	9	81.8	10	58.8	

Markedly improved	1	9.1	2	11.8	
Not determined	0	0	0	0	
Total	11	100	17	100	-
12 weeks					
Worsened	0	0	0	0	
No change	3	27.3	4	23.5	
Improved	4	36.4	4	23.5	0.7005
Markedly improved	4	36.4	9	52.9	
Not determined	0	0	0	0	
Total	11	100	17	100	-

Table 12: Patients' impression of treatment effect [Comparison between groups using Fisher's exact test].

When steroids are administered by local injection, Samrao et al. recommended monitoring the bone mineral density (BMD) of AA patients to avoid the risk of steroidal osteoporosis because they found abnormal BMD in 50% of patients after 20 weeks or longer treatment with triamcinolone acetonide in 4-8 week cycles and because the cumulative dose of triamcinolone acetonide is a risk factor for steroidal osteoporosis [18]. On the other hand, it was reported that glycyrrhizic acid prevents steroid-induced osteoporosis in rats [19]. Glycyrrhetinic acid was reported to inhibit 11β-HSD1 of the enzyme metabolizing cortisone to cortisol [20], and thus it was inferred that glycyrrhetinic acid prevents osteoporosis by promoting the metabolism of cortisol produced in excess due to steroid treatment. In other words, glycyrrhetinic acid acts on both the enzyme that activates and the enzyme that inactivates cortisol and plays a role in the treatment of AA by promoting cortisol production when the response to corticotropin-releasing hormone is decreased. Glycyrrhetinic acid also prevents side effects by increasing the metabolism of excess cortisol produced in response to steroid administration.

Average area (cm^2)														
	n	Start			4 weeks			8 weeks			12 weeks			
Single treatment	11	1.85	±	1.22	1.44	±	1.15	1.08	±	0.95	0.89	±	1.24	
p-value (within group)*					0.1581			0.126			0.0966			
Combined treatment	17	2.63	±	2.78	2.06	±	2.54	1.56	±	2.25	0.95	±	1.54	
p-value (within group)*					0.0387			0.0078			0.0009			
p-value (between groups)**		0.7419			0.7598			0.9064			1			
Reduction rate (%)														
Single treatment					-20.84	±	29.56	-35.88	±	40.64	-46.64	±	55.84	
Combined treatment					-15.83	±	41.91	-45.79	±	37.25	-61.12	±	48.57	
p-value (between groups)**					0.8323			0.4659			0.6592			

*Comparison wihin each group using the Bonferroni multiple procedure.

**Comparison between groups using Wilcoxon's exact test.

Table 13: Change in area (area observed) of hair loss.

Since it has been recognized that the serum oxidative stress marker level is higher and the antioxidative stress marker is lower in AA patients in than in healthy people, it was suggested that oxidative stress is involved in the development of the condition [21,22]. Glycyrrhizic acid is known to exert antioxidative effects [23-25], suggesting that it could improve symptoms by decreasing oxidative stress in AA.

CC hydrate, the active ingredient in CC solution, has local vasodilator activity, increasing blood flow in capillaries and promoting local metabolism by stimulating acetylcholine receptors of vascular smooth muscle. CC hydrate promotes hair growth by acting on degraded hair follicles [26,27].

Among the various treatments for AA, steroids have been administered most frequently because their mechanism of action is

clear. However, many patients have reservations about steroid treatment even under the supervision of a physician. MG tablets are viewed as safe in terms of both lack of side effects and treatment efficacy and they contribute to the maintenance of homeostasis of corticosteroids in hair follicle tissue.

Average area (cm^2)													
	n	Start			4 weeks			8 weeks			12 weeks		
Single treatment	6	2.11	±	1.52	1.63	±	1.53	1.3	±	1.26	1.17	±	1.62
p-value (within group)*					0.6564			0.4689			0.6564		
Combined treatment	7	2.95	±	3.84	2.34	±	3.45	1.27	±	2	0.46	±	0.69
p-value (within group)*					0.3282			0.0468			0.0468		
p-value (between groups)**		1			0.6682			0.5677			0.7072		
Reduction rate (%)													
Single treatment					-22.62	±	34.48	-41.53	±	35.08	-45.32	±	66.18
Combined treatment					-26.84	±	30.3	-65.80	±	20.02	-86.76	±	13
p-value (between groups)**					1			0.0865			0.4989		
*Comparison within each group using the Bonferroni multiple procedure. **Comparison between groups using Wilcoxon's exact test.													

Table 14: Change in area (area observed) of hair loss in patients with atopic predisposition.

The limitations of this study were, because it was an interventional comparison, the number of patients differed between the two groups, and the overall sample size was small. Therefore, no statistically significant difference was seen between groups. Since the present combination therapy obviously showed the decrease in the area of AA and the effects in patients with allergic factors, we proceed to further larger randomized trials to confirm the results in this study.

In conclusion, in the present interventional study, MG tablets combined with CC solution therapy showed decreases in areas of AA without serious adverse events, especially in patients with allergic factors.

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