

Comparative Study of Efficacy and Safety of Topical Squaric Acid Dibutylester and Diphenylcyclopropenone for the Treatment of Alopecia Areata

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Abstract

Background: Topical squaric acid dibutylester and diphenylcyclopropenone are still the most effective therapy for alopecia areata among widely available treatment options. Hence, it is important to know which one is more effective and safer between the two. **Aims:** The aim of this study was to compare topical squaric acid dibutylester and diphenylcyclopropenone for the treatment of alopecia areata in terms of their efficacy and side effects. **Subjects and Methods:** In the time period of January–March 2015, a total of 40 patients were selected for this study from the outpatient department of Rajendra Institute of Medical Sciences, Ranchi. After dropout of 16 patients, the remaining 24 patients were randomly divided into two groups; that is, group A for squaric acid dibutylester and group B for diphenylcyclopropenone. Each group received treatment for 6 months between March–November 2015. Their efficacy and side effects were compared. **Statistical Test:** Unpaired student *t*-test was performed. $P < 0.05$ was considered to be significant and 95% confidence interval was also used to evaluate the efficacy. **Results:** The mean values of percentage change in baseline severity of alopecia tool score for squaric acid dibutylester and diphenylcyclopropenone were 52.25 and 34.45, respectively. At 6 months, 95% confidence interval was 43.5–61% for group A and 25–44% for group B. In 58.33% of group A patients, A3 (50–74%) grade of improvement was observed, whereas in group B patients, it was 33.33%. A4 grade of improvement (75–99%) was also seen in 1 patient of group A. Minor side effects were seen in 2 patients of group A and 10 patients of group B. None of the group A patients showed major side effects, however, 2 patients suffered major side effects in group B. **Conclusions:** Between squaric acid dibutylester and diphenylcyclopropenone, squaric acid dibutylester is more efficacious. Further, frequencies of major and minor side effects are also lower than diphenylcyclopropenone.

Keywords: Alopecia areata, diphenylcyclopropenone, squaric acid dibutylester

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Introduction

Alopecia areata (AA) is a chronic inflammatory, recurrent, tissue specific autoimmune disease, mediated by autoreactive CD8+ T cells, occurring in genetically predisposed individuals and characterized by sudden

or gradual onset, presenting with oval or round, well-circumscribed, nonscaly and nonscarring bald patches with smooth surface on the scalp or elsewhere

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on the body.^[1,2] Its worldwide prevalence is 0.2% and the lifetime risk is 2%.^[3-5] There is no sex predilection.^[3] Positive family history is usually seen in 10–42% of the cases.^[2,6] At some point of time, 5% cases of AA may turn into alopecia totalis and 1% of the cases end up as alopecia universalis.^[1,2,4]

A number of treatment modalities have been tried in topical, oral, and injection formulations to induce hair growth in alopecia areata, however, none has shown permanent alteration of the long-term course of the disease.^[1,2] Although pathogenesis of AA is yet to be completely elucidated, taking the best current knowledge of pathogenesis of AA into consideration, targeting intrabulbar and peribulbar lymphocytic infiltrates by using squaric acid dibutylester (SADBE) and diphenylcyclopropenone (DPCP) in contact immunotherapy is by far the best chemotherapy for AA.^[7-9] Since 1980, many studies have been conducted to determine the efficacy and safety of SADBE and DPCP individually, however, to the best of our knowledge, none of the studies have been conducted to compare these two chemicals' efficacy and side effects in the same sitting. Here, we investigate and compare the efficacy and side effects of SADBE and DPCP in patients of AA in a randomized, single blinded, uncontrolled, prospective study.

Subjects and Methods

Study design

A randomized, single blinded, uncontrolled, prospective study was conducted among 24 patients of AA to compare 6-month treatment with topical SADBE and DPCP. Randomly allocated group A and group B, each with 12 patients of AA, were treated with SADBE and DPCP, respectively. The study was conducted in accordance with the Declaration of Helsinki and good clinical practice guidelines. The protocol of treatment was approved by the institutional review board and informed consent was obtained from all patients.

Study area

Department of Dermatology, Venereology and Leprosy, Rajendra Institute of Medical Sciences, Ranchi, India.

Study population

A total of 40 patients of AA were enrolled from the outpatient department, out of which 24 patients were available for the study due to dropout of 16 cases.

Patient's selection

On the basis of history and clinical examination, diagnosis of AA was made, and the following criteria were used during the selection of the patients:

Inclusion criteria

- Immunocompetent patients
- Patients willing for regular follow-up after taking consent for treatment
- Patients >18 years of age
- Alopecia areata affecting >20% of scalp area
- No local/systemic therapy since at least 6 months.

Exclusion criteria

- Patients not willing for treatment and/or regular follow-up
- Pregnancy
- Serious intercurrent medical illness
- Significant cardiovascular disease
- Alopecia areata affecting <20% of scalp area
- Localized scalp infections/inflammation
- Immunosuppressed patients
- History of local/systemic therapy within 6 months.

A detailed history, general physical examination, and local examination were conducted for each patient and recorded on a proforma as per the guidelines of National Alopecia Areata Foundation (NAAF).^[10,11] Following laboratory tests were performed for each patient:

Laboratory tests

- Complete blood count
- Routine examination of urine
- Blood sugar
- Renal function test
- Liver function test
- Thyroid function test
- Total serum IgE
- ANA and Anti ds DNA
- HIV
- VDRL/RPR for syphilis
- KOH smear and fungal culture
- Light microscopy.

Study treatment

The application of contact allergens involved two phases:
A. Sensitization phase
B. Elicitation phase

Sensitization

Initially, 2% concentration was applied with a cotton-tipped applicator to a 4 cm × 4 cm square area on the bald patch of the scalp to sensitize the patient [Figure 1]. Patients were advised to wait for 5–10 min and then cover their head with a cap or cloth to protect it sunlight and not to wash their scalp for 48 h after application. After 48 h, patients were also advised to shampoo the scalp to remove the residual allergens.

Elicitation

- 2 weeks later, a very lower concentration of 0.001% was applied on the same bald patch or the same side of the scalp (if both sides are involved). The patients were advised to take the same precautions after the application
- On a weekly basis, concentration of the allergen was increased very gradually as 0.01%, 0.05%, 0.1%, 0.5%, 1%, etc., until a mild dermatitis manifesting as low-grade erythema and mild pruritus were obtained which persisted for 48 h. Then, this concentration was applied weekly
- Usually, between 0.01–1% desired contact dermatitis was observed to occur. After establishing the appropriate concentration, therapy was continued on a weekly basis for 6 months
- Once hair regrowth was complete and maintained for more than 3 months, treatment was tapered in the same manner and discontinued over a period of 9 months
- If there was no response in 6 months, immunotherapy was stopped and the patient was considered as “non-responder.”

Precautions

1. Contact immunotherapy is an unlicensed treatment that uses a nonpharmaceutical grade agent
2. Patients should
 - a. be fully informed about the nature of the treatment,
 - b. be given an information sheet, and
 - c. they should provide signed consent.
3. Dilution should always be done in acetone
4. Solution should strictly be stored in air tight, screw-capped, amber-coloured containers in a dark room
5. Storage of the diluted solution is difficult because acetone often evaporates, leading to a change in



Figure 1: Application on 4 cm × 4cm area on a bald patch of the scalp

- the concentration. Therefore, ideally, fresh solution should be made every time to avoid this
6. Application needs to be done fast to cover the entire scalp before acetone starts evaporating in the beaker
 7. Great care must be taken to avoid contact with the allergen by handlers, including pharmacy, medical, and nursing staff, as well as other members of the patient’s family. Those applying the allergen should wear gloves and aprons
 8. Patients should be advised to wait for 5–10 min before covering their head with a cap or cloth to protect from sunlight
 9. Patients are advised not to wash their scalp for 48 h after DPCP application. After 48 h, patients should be advised to shampoo the scalp to remove the residual DPCP. During these 48 h, patients should be advised to avoid touching the scalp accidentally either by themselves or by others
 10. There is no data on the safety of contact immunotherapy during pregnancy and it should neither be used in pregnant women nor in women intending to become pregnant.

Assessment of the extent of hair loss by Severity of Alopecia Tool score scoring

NAAF working committee has devised Severity of Alopecia Tool score (SALT) score.^[12] The scalp is divided into 4 areas, namely, vertex: 40% (0.4) of scalp surface area; right profile of scalp: 18% (0.18) of scalp surface area; left profile of scalp: 18% (0.18) of scalp surface area; and posterior aspect of scalp: 24% (0.24) of scalp surface area [Figure 2]. Percentage of hair loss in any of these areas is percentage hair loss multiplied by

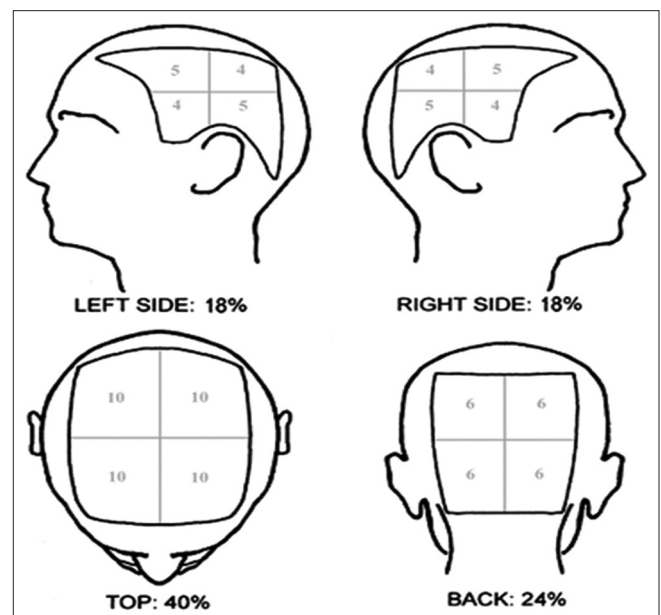


Figure 2: Calculation of extent of hair loss by SALT scoring

percent surface area of the scalp in that area. SALT score is the sum of the percentage of hair loss in all the abovementioned areas. For example, if the percentage hair loss in vertex, right profile, left profile, and posterior aspect is 20, 30, 40, and 50% respectively; then, SALT score = $(20 \times 0.4) + (30 \times 0.18) + (40 \times 0.18) + (50 \times 0.24) = 8 + 5.4 + 7.2 + 12 = 32.6$.

Assessment of the efficacy of therapy

Efficacy of SADBE and DPCP was assessed and compared on the basis of absolute change in the SALT score and the percentage hair regrowth derived from change in baseline SALT score.

Absolute change in SALT score = SALT score at baseline - SALT score at 24 weeks.

Percent scalp hair re-growth based on SALT score = $100 \times (\text{Baseline SALT score} - \text{SALT score at 24 weeks}) / \text{Baseline SALT score}$.

Assessment of percentage hair regrowth was graded into following 6 grades:

- A0 = no change or further loss of hairs
- A1 = 1-24% regrowth
- A2 = 25-49% regrowth
- A3 = 50-74% regrowth
- A4 = 75-99% regrowth
- A5 = 100% regrowth.

Statistical evaluation

For this study, analytical and descriptive evaluations were performed. To compare the efficacy of SADBE and DPCP in the treatment of alopecia areata, unpaired student *t*-test was performed with the help of IBM SPSS version 22.0. (Armonk, NY, IBM corp.) data software. The mean scores of both the groups were compared to calculate the level of significance (*P* value). *P* value was assessed at 5% and *P* < 0.05 was considered to be significant. Side effects profile of SADBE and DPCP were evaluated only descriptively.

Results

Patient’s demographics

Most of the patients were in the age group of 21-30 in both the groups. Male:Female ratio for group A was 1:1 and for group B it was 2:1.

Baseline characteristics of alopecia areata in both groups

In group A, 58.33% had multiple patches of alopecia areata. Ophiasis and patchy (single) variety accounted for 16.67% each. In group B, 50% of the cases had multiple patches of alopecia areata and 25% had a single patch. Ophiasis and

reticular types of alopecia areata consisted of 16.67 and 8.33%, respectively. Most of the cases in both the groups belonged to the subacute category. Group A had 83.34% and group B had 75% cases in the subacute category.

Changes in Severity of Alopecia Tool score after treatment

Absolute change in the baseline SALT score was calculated. Mean value of the absolute change in SALT score for group A patients was 21.33 and for group B patients was 14.08.

Percentage scalp hair regrowth was derived from the absolute change in the baseline SALT score for all the patients. The mean values were 52.25% for group A and 34.45% for group B [Table 1].

In group A, 58.33% cases showed A3 grade of improvement, and A2 grade was seen in 33.33%. In group B, A2 grade of improvement was seen in 41.67% and A3 in 33.33%. One case showed no response at all [Table 2].

Table 1: Percentage changes in baseline SALT score in group A and B patients (percentage scalp hair regrowth)

Patients	Percentage change in baseline SALT score		Percentage scalp hair growth	
	A	B	A	B
Patient 1	75	16.66	75	16.66
Patient 2	63.66	50	63.66	50
Patient 3	30	33.33	30	33.33
Patient 4	57.14	42.85	57.14	42.85
Patient 5	40	25	40	25
Patient 6	50	55.55	50	55.55
Patient 7	25	40	25	40
Patient 8	60	16.66	60	16.66
Patient 9	66.66	0	66.66	0
Patient 10	66.66	50	66.66	50
Patient 11	50	33.33	50	33.33
Patient 12	42.85	50	42.85	50
Mean	52.25	34.45	52.25	34.45

SALT = Severity of Alopecia Tool score

Table 2: Grading of overall improvement in groups A and B

Overall improvement	Group A	Group B	Percentage	
			A	B
A0 (no hair regrowth)	0	1	0	8.33
A1 (1-24%)	0	2	0	16.67
A2 (25-49%)	4	5	33.33	41.67
A3 (50-74%)	7	4	58.33	33.33
A4 (75-99%)	1	0	8.33	0
A5 (100%)	0	0	0	0
Total	12	12	100	100

Side effects profile

Minor side effects were seen in 10 patients of group B and 2 patients of group A. Regional lymphadenopathy was the most commonly observed side effect seen in 16.66% of group A and 33.33% of group B patients. None of the group A patients had major side effects whereas in group B it was seen in 2 patients [Figure 3].

P value

With the unpaired student *t*-test, *P* value was calculated by computing the two mean values of percentage scalp hair regrowth in both the groups; it was calculated to be 0.014, which is significant enough in favor of group A patients' treatment with SADBE.

Discussion

The severity of AA is variable and the clinical hallmark is nonscarring, patchy hair loss, which may recover spontaneously; however, rarely it may progress to long lasting chronic alopecia totalis or universalis.^[1,2] The pathogenesis of AA is still uncertain, however, now there are many indirect evidences that signify the importance of CD4+ and CD8+ T cells and cytokines such as IL-1 α , IL-1 β , IL-2, IL-10, IL-17, IFN- γ , and TNF- α in its pathogenesis.^[13,14] Owing to its autoimmune basis, several types of treatment modalities have been tried in different studies but none of them has been found to be a definitive and curative treatment. Contact immunotherapy is the most effective and best-documented treatment for alopecia areata so far.^[15,16] At present, SADBE and DPCP are used as contact allergens of topical immunotherapy. Despite being used since the last 35 years, there are not many studies conducted worldwide to compare the

efficacy and side effects of SADBE and DPCP. At present, DPCP has been considered as the "contact allergen of choice" because it is more stable in acetone and is relatively cheaper than SADBE.^[17] Poor accessibility, high cost of the drug, and poor patient compliance are major limiting factors of this therapy.^[18] There are also some poor prognostic factors such as early age of onset, chronicity of the disease, ophiasis, AT/AU, personal/family history of other autoimmune disease, nail changes, and atopy.^[18]

In this study, patients of group A were treated with SADBE and group B with DPCP. Mean value of absolute change in the SALT score was more for group A patients (21.33) than that for group B patients (14.08). Mean value of percentage change in the baseline SALT score for group A and B patients were 52.25 and 34.45, respectively [Figure 4]. At 6 months, 95% confidence interval was calculated to be 43.5–61% for group A and 25–44% for group B.

In 58.33% of group A patients, A3 grade of improvement was observed whereas in group B patients, it was only 33.33%. A4 grade of improvement was also seen in 1 patient of group A. None of the patients of group A were nonresponders whereas one of the group B patients could not show any hair growth [Table 2].

Frequencies of major and minor side effects were more in group B patients treated with DPCP.

P value was calculated to be 0.014 by comparing the mean values of percentage change in the baseline SALT score of both the groups. *P* < 0.05, which was significant enough to consider SADBE to be more efficacious than DPCP.

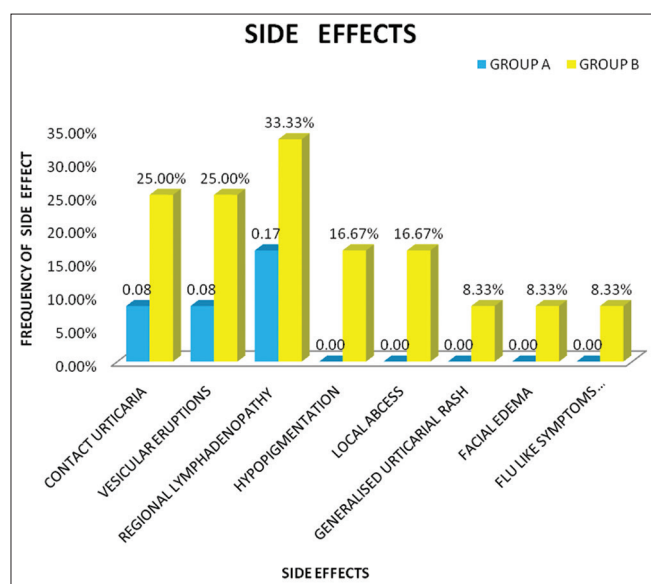


Figure 3: Frequencies of major and minor side effects in both groups

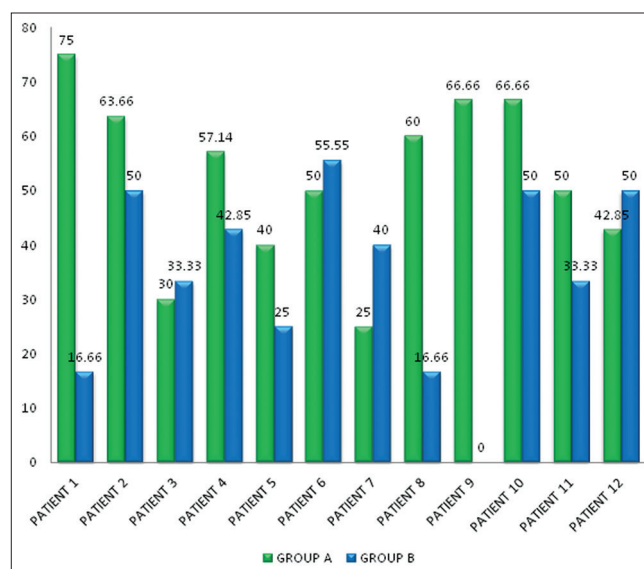


Figure 4: Percentage change in SALT score and percent scalp hair regrowth in patients of groups A and B

To the best of our knowledge, there are no previously conducted studies comparing the efficacy of SADBE and DPCP at the same time. Individual studies of their efficacy have been conducted separately time and again. Other studies have compared these chemicals by assessing their response one at a time in different time-periods separated by a gap period. Parallel study assessing them together in the same time period has not been conducted till date to the best of our knowledge.

Conclusion

To conclude, topical immunotherapy was an effective treatment modality for the patients of alopecia areata, and between the two currently available contact sensitizers, SADBE, and DPCP, SADBE was more efficacious than DPCP and frequencies of both, major, and minor side effects with SADBE were also lower than with DPCP.

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Nil.

Conflicts of interest

There are no conflicts of interest.

References

- Gilhar A, Kalish RS. Alopecia areata: A tissue-specific autoimmune disease of the hair follicle. *Autoimmun Rev* 2006;5:64-9.
- Amin SS, Sachdeva S. Alopecia areata: An update. *J Pakistan Assoc Dermatol* 2013;23:209-20.
- Safavi KH, Muller SA, Suman VJ, Moshell AN, Melton LJ 3rd. Incidence of alopecia areata in Olmsted County, Minnesota, 1975 through 1989. *Mayo Clin Proc* 1995;70:628-33.
- SharmaVK, Dawn G, Kumar B. Profile of alopecia areata in Northern India. *Int J Dermatol* 1996;35:22-7.
- Thappa DM, Vijayikumar M. Alopecia areata. *Indian J Dermatol Venereol Leprol* 2001;67:188.
- Goh C, Finkel M, Christos PJ, Sinha AA. Profile of 513 patients with alopecia areata: Associations of disease subtypes with atopy, autoimmune disease and positive family history. *J Eur Acad Dermatol Venereol* 2006;20:1055-60.
- Happle R, Klein H, Macher E. Topical immunotherapy changes in the composition of the peribulbar infiltrate in alopecia areata. *Arch Dermatol Res* 1986;278:214-18.
- Happle R, Kalveram KJ, Büchner U, Echternacht-Happle K, GöggeImann W, Summer KH. Contact allergy as therapeutic tool for alopecia areata: Application of squaric acid dibutylester. *Dermatologica* 1980;161:289-97.
- Gianetti A, Orecchia G. Clinical experience on the treatment of alopecia areata with squaric acid dibutylester. *Dermatologica* 1983;167:280-2.
- Olsen EA, Hordinsky MK, Price VH, Roberts JL, Shapiro J, Canfield D, *et al.* Alopecia areata investigational assessment guidelines. Part II. National Alopecia Areata Foundation. *J Am Acad Dermatol* 2004;51:440-7.
- Olsen E, Hordinsky M, McDonald-Hill S, Price V, Roberts J, Shapiro J, *et al.* Alopecia areata investigational assessment guidelines. *J Am Acad Dermatol* 1999;40:242-6.
- Olsen EA, Hordinsky MK, Price VH, Roberts JL, Shapiro J, Canfield D, *et al.* 2004. National alopecia areata foundation.
- Freyschmidt-Paul P, Happle R, Hoffman R. Alopecia areata in animal models—new insights into pathogenesis and treatment of a T cell-mediated autoimmune disorder. *J Dtsch Dermatol Ges* 2004;2:260-73.
- Philpott MP, Sanders DA, Bowen J, Kealey T. Effects of interleukines, colony-stimulating factor and tumor necrosis factor on human hair follicle growth *in vitro*: A possible role for interleukin-1 and tumor necrosis factor- α in alopecia areata. *Br J Dermatol* 1996;135:942-8.
- Case PC, Mitchell AJ, Swanson NA, Vanderveen EE, Ellis CN, Headington JT. Topical therapy of alopecia areata with squaric acid dibutylester. *J Am Acad Dermatol* 1984;10:447-50.
- Dall’oglio F, Nasca MR, Musumeci ML, La Torre G, Ricciardi G, Potenza C, *et al.* Topical immunomodulator therapy with squaric acid dibutylester (SADBE) is effective treatment for severe alopecia areata (AA): Results of an open-label, paired-comparison, clinical trial. *J Dermatolog Treat* 2005;16:10-4.
- Wilkerson MG, Henkin J, Wilkin JK, Smith RG. Squaric acid and esters: Analysis for contaminants and stability in solvents. *J Am Acad Dermatol* 1985;13:229-34.
- Van der Steen PH, Van Baar HM, Happle R, Boezeman J, Perret CM. Prognostic factors in the treatment of alopecia areata with diphenylcyclopropanone. *J Am Acad Dermatol* 1991;24:227-30.