

Double-blind, randomized, placebo-controlled study of a lotion containing triethyl citrate and ethyl linoleate in the treatment of acne vulgaris

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Summary

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Key words

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Conflicts of interest

None declared.

Background Acne vulgaris is a major clinical problem; despite a vast array of treatment modalities available for acne, there is considerable dissatisfaction in acne treatment among patients and doctors. Rising antibiotic drug resistance consequent to the widespread use of topical antibiotics is causing concern and effective nonantibiotic treatments are needed.

Objectives To evaluate the efficacy and tolerability of a novel lotion containing triethyl citrate and ethyl linoleate in the treatment of mild to moderate acne vulgaris.

Methods This was a double-blind, placebo-controlled, randomized study comparing the active lotion containing triethyl citrate and ethyl linoleate with its vehicle as a placebo control. Patients were assessed by the modified Leeds acne grading system as well as by counting inflammatory and noninflammatory lesions on the face at weeks 0, 4, 8 and 12. Sebum production was assessed by the Sebutape method at weeks 0 and 12. All adverse events were recorded.

Results Forty patients were recruited into the study, of whom 33 completed the study. Active treatment was statistically superior to placebo in reduction of Leeds grading and total, inflammatory and noninflammatory lesion counts. The active lotion showed a rapid response with obvious reduction in lesion counts and acne grading by 4 weeks. Sebum production was significantly reduced in the actively treated group, with a mean reduction of 53% in sebum production compared with baseline. One patient developed irritation to the active lotion and withdrew from the study.

Conclusions The new lotion containing triethyl citrate and ethyl linoleate has been shown to be an effective treatment for mild to moderate acne, with an effect on both inflammatory and noninflammatory acne lesions. The new lotion worked quickly and was generally well tolerated. A surprising finding was the significant impact the new lotion has on sebum production, suggesting a role in patients with seborrhoea. This nonantibiotic preparation will be a very useful addition to existing treatments for acne.

Acne vulgaris is the most common chronic inflammatory disease of the skin to affect humans. It affects all ages but mainly young people at the sensitive period of puberty and can have an adverse effect on their psychological development, which may lead to social phobias, withdrawal from society and clinical depression.¹⁻³

Various factors contribute to the pathogenesis of acne, including increased androgen-mediated sebum production, alteration of sebum composition, hyperproliferation of the follicular keratinocytes and colonization of the pilosebaceous

duct with *Propionibacterium acnes*. Current treatments for acne include topical and oral antibiotics, topical antimicrobials and topical and oral retinoids. All acne treatments have potential side-effects, some of which may be severe. Topical treatments and oral antibiotics generally need to be used for several months to achieve a response, which leads to major problems with patient compliance. In addition to possible side-effects, long-term exposure to antibiotics has exerted enormous selective pressure on the bacterial skin flora of patients with acne, with the emergence of antibiotic-resistant

propionibacteria.⁴⁻⁷ The increasing number of failures with classic treatments has emphasized the need to develop new therapeutic options for the treatment of acne which are preferably nonantibiotic.

In this study we have investigated the efficacy and tolerability of a novel topical lotion composed of triethyl citrate and ethyl linoleate (AkmicareTM lotion; SkinMed, Harrogate, U.K.) as the active agents in the treatment of mild to moderate acne vulgaris.

Patients and methods

This was a double-blind, randomized, vehicle-controlled study comparing a lotion containing triethyl citrate and ethyl linoleate and the vehicle in the treatment of mild to moderate acne vulgaris. Ethics committee approval was granted to conduct the study.

Study population

Individuals were recruited from those attending the dermatology clinic at the Hammersmith Hospital, with written informed consent. Inclusion criteria were patients aged between 16 and 45 years with mild to moderate facial inflammatory acne defined as the presence of at least 10 acne papules or pustules between the brow and jaw line and an acne severity score of between 2 and 7 on the Leeds revised acne grading system.⁸ Exclusion criteria included severe acne, rosacea, pregnancy, breastfeeding, known allergy to constituents of the lotions, use of medication for acne or use of antibiotics for other medical conditions. Patients who were receiving therapy for acne and were willing to participate in the trial entered a washout period, the duration of which depended on the treatment (4 weeks for oral and topical antibiotics, 12 weeks for cyproterone acetate-containing contraceptives and 52 weeks for oral isotretinoin).

Study protocol

At recruitment, a short medical history including demographic details and clinical assessment with acne grading and total lesion counts (inflammatory lesions – papules and pustules, and noninflammatory lesions – open and closed comedones) were recorded for each patient.⁸ Patients were randomized to treatment with active lotion or vehicle by a computer-generated sequence. The two lotions were provided in identical bottles with applicator tips labelled A or B, ensuring anonymity of the product for both the investigator and the patient (double-blind randomized trial). The patients were treated with the active lotion or the vehicle twice daily for 3 months and were reviewed at 4, 8 and 12 weeks. At each visit any side-effects were recorded and patients were clinically assessed for acne grading and accurate spot counts were performed. Sebum production was documented at the first and last visit.

Acne evaluation

To evaluate acne the Leeds revised acne grading system was used. This is a rapid and reproducible means of recording inflammatory acne by matching acne severity with validated photographs of patients with acne and assigning a numerical score between 1 and 12. This technique provides a straightforward means of clinical acne classification and has become established as a grading method in many clinical trials of acne treatment.⁸ Lesion counts were recorded for the whole face (excluding the nose) by individually counting noninflammatory (blackheads and whiteheads) and inflammatory (papules, pustules, nodules and cysts) lesions. All adverse events were assessed by direct questioning of patients.

Sebum assessment

Sebum excretion rate was measured at week 0 and week 12 using a Sebutape (sebum-absorbent tape technique). The forehead was cleaned with soap and was then defatted by wiping with gauze soaked in hexane. The sebum-absorbent tape was applied to the central forehead and was removed 1 h later. Quantitative analysis was performed with photometric techniques as previously described.⁹

Study endpoint

The primary endpoints of the study were change in acne severity and sebum production after 12 weeks based on the Leeds revised grading system and the photometric measurements of the Sebutape, respectively, and adverse events at any time. Secondary endpoints were changes in total, inflammatory and non-inflammatory lesion counts by the end of the trial.

Statistical analysis

We analysed our data by using the statistical package SPSS 11 (SPSS, Chicago, IL, U.S.A.) using an intention-to-treat and per-protocol basis analysis. We calculated that 15 people in each group would have the power to demonstrate a 50% difference in the primary endpoints relative to their baseline measurement with 90% power and at 5% significance level. To insure for possible dropouts we increased our population in each group by 33%. All values were expressed as median (interquartile range). Non-normally distributed data were analysed using the Mann–Whitney U-test for independent groups. We used a linear regression model to assess the impact of different variables on inflammatory, noninflammatory and total lesion count, acne grade and sebum production. We fitted a forward stepwise multivariate regression model using the following baseline characteristics that were judged capable of affecting outcome: age, sex, age at onset of acne, duration of acne, skin type, previous use of oral isotretinoin, and previous use of oral antibiotics for acne. On regression, any variable that was judged unimportant ($P > 0.25$) was discarded from the multivariate analysis.

Results

Forty patients were recruited in the study and were assessed by two investigators. At the first visit all patients were assessed by both investigators. Thereafter, where possible both investigators assessed. Twenty patients received the active treatment and 20 the vehicle. Three of 20 patients (15%) on active treatment withdrew: one left the locality, the second developed depression considered unrelated to the treatment and the third patient developed a skin rash. Four of 20 patients treated with vehicle (20%) withdrew by 4 weeks because of dissatisfaction with clinical response.

Table 1 shows the baseline demographic and clinical characteristics of the two groups. Most patients were young adults who had a long history of acne. A similar proportion of

Table 1 Patients' characteristics. Data are median (interquartile range) or number (%)

	Active group	Placebo group
Age (years)	24 (20–30.75)	27.5 (18.25–33)
Sex: M/F	7 (35%)/13 (65%)	11 (55%)/9 (45%)
Age at onset of acne (years)	15 (14–17.75)	15.5 (14–16)
Duration of acne (years)	6.75 (3.5–16)	10.25 (5–17.75)
Skin type (Fitzpatrick classification)	4 (2–5)	3 (2–4.75)
Ethnic origin		
White	14 (70%)	15 (75%)
Asian	4 (20%)	3 (15%)
Afro-Caribbean	2 (10%)	2 (10%)
Previous treatments		
Oral antibiotics	13 (65%)	14 (70%)
Oral isotretinoin	8 (40%)	7 (35%)
Topical retinoids	15 (75%)	16 (80%)
Topical benzoyl peroxide	15 (75%)	16 (80%)

Table 2 Acne severity, lesion counts and sebum production at baseline and 12 weeks after treatment. Data are shown as median (interquartile range)

	Baseline	n	12 weeks	n	P-value
Severity (Leeds revised grade)					
Active	4 (3–5)	20	2 (1–2.75)	20	0.001
Placebo	4 (3–5)	20	5 (3.25–6)	20	
Inflammatory lesions					
Active	18.5 (15–24.5)	20	8 (4.25–16)	20	0.001
Placebo	20 (17.25–28.5)	20	28.5 (19–34)	20	
Noninflammatory lesions					
Active	31.5 (19.25–45.25)	20	15 (6.25–24.75)	20	0.001
Placebo	27.5 (17.75–43.75)	20	44.5 (28–64)	20	
Total lesions					
Active	49 (40.5–64.5)	20	25 (12.25–34.5)	20	0.001
Placebo	52 (37.25–82.5)	20	78.5 (48–104)	20	
Sebum production					
Active	91.93 (38.07–135.81)	20	42.9 (18.65–95.61)	20	0.001
Placebo	86.32 (22.11–105)	20	107.59 (58.57–131.28)	20	

patients in each group had previously received systemic antibiotics or oral isotretinoin and all ethnicity groups were represented in both groups.

Table 2 shows the clinical acne assessment and sebum production of the two groups of patients. The two groups were well matched at the start of the study, with no significant differences in acne grade, inflammatory lesion count, noninflammatory lesion count or sebum production.

At 12 weeks marked improvement in acne grade, inflammatory lesion count, noninflammatory lesion count, total lesion count and a reduction in sebum production were observed in the actively treated group. All parameters were significantly better than those of the vehicle-treated group. Improvement in acne grade and reduction of lesion counts were observed as early as 4 weeks in the actively treated group. Sebum production fell by a mean of 53% (range 35–68%) by the end of the study.

Forward stepwise regression analysis failed to identify any factor other than treatment-group allocation that substantially affected the reduction in acne severity by 12 weeks (Table 3).

Figures 1–4 show the observed mean change in inflammatory lesion count, noninflammatory lesion count, overall acne severity and total lesion count throughout the trial. Figure 5 shows the effect of the treatment on sebum production. Rapid improvement in the lesion counts and the acne severity occurred in the first 4 weeks of treatment with the active lotion.

Adverse events were reported in only one patient in the active treatment group, who developed erythema, irritation and desquamation, and the treatment was stopped. No patients in the vehicle-treated group reported adverse events.

Discussion

This study demonstrates that this new lotion containing triethyl citrate and ethyl linoleate (Akmicare™) is a very effective and well-tolerated topical agent in the treatment of both acne and seborrhoea. The use of lotion twice daily for 12 weeks was associated with significant improvement in acne severity

Table 3 Regression analysis to assess effect of baseline characteristics on improvement of acne severity

	Coefficient value (r)	95% CI	P-value
Age	-0.013	-0.066 to -0.040	0.62
Treatment group	-1.461	-2.178 to -0.744	
Sex	0.289	-0.443 to 1.022	0.43
Treatment group	-1.392	-2.121 to -0.663	
Age at onset of acne	-0.063	-0.169 to 0.043	0.24
Treatment group	-1.422	-2.133 to -0.710	
Duration of acne	0.000	-0.004 to 0.005	0.88
Treatment group	-1.444	-2.166 to -0.731	
Use of oral antibiotics	0.029	-0.738 to 0.795	0.94
Treatment group	-1.449	-2.166 to -0.731	
Use of oral isotretinoin	0.171	-0.569 to 0.911	0.65
Treatment group	-1.459	-2.175 to -0.742	

CI, confidence interval.

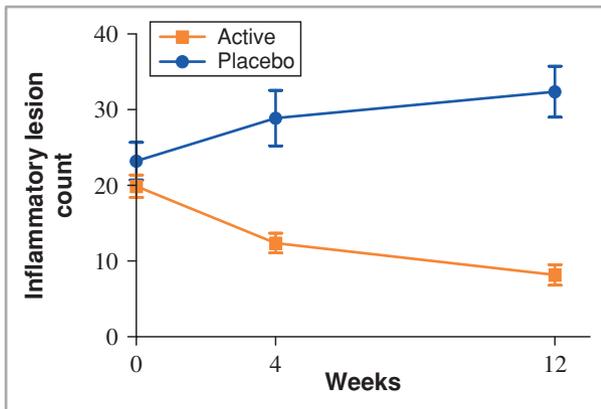


Fig 1. Change in inflammatory lesion count with time.

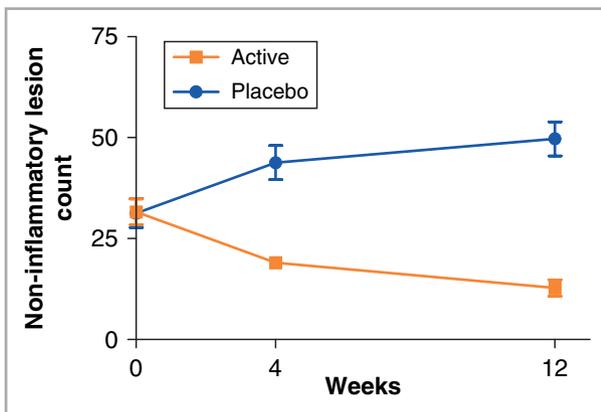


Fig 2. Change in noninflammatory lesion count with time.

compared with the vehicle. This improvement was seen for a range of disease severity and included complete clearance in two patients. The reduction in severity was indicated by a corresponding fall in total, inflammatory and noninflammatory lesion counts. In addition, sebum excretion was

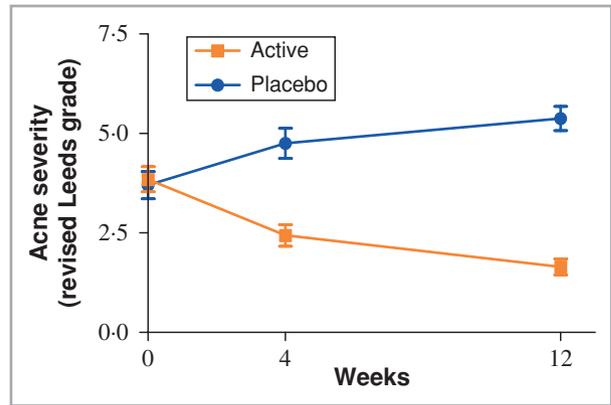


Fig 3. Change in acne severity with time.

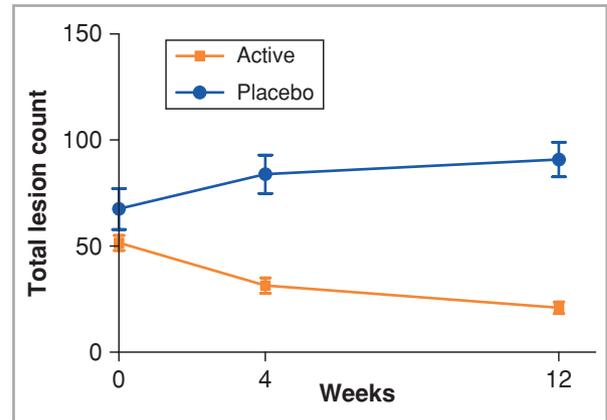


Fig 4. Change in total lesion count with time.

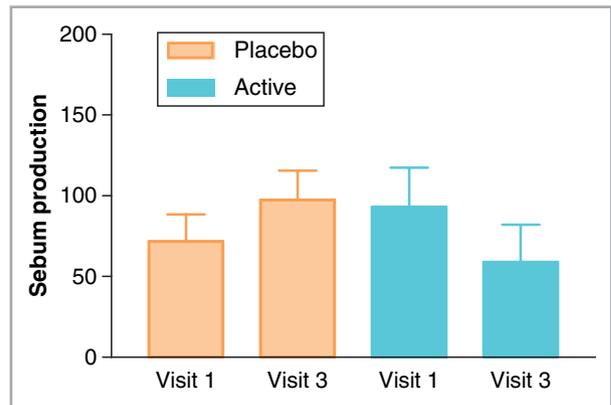


Fig 5. Mean \pm SD sebum production at baseline (visit 1) and 12 weeks (visit 3).

reduced by up to 68%. The results were apparent even from the first 4 weeks of treatment and the rapidity of the response to this lotion contrasts with that to conventional treatments such as oral antibiotics, that often need administration for 6–8 weeks before benefits are seen.

In our study, the sebum excretion rate was measured using Sebutape. The limitation of the use of Sebutape as a method

for the evaluation of sebum production is that a number of environmental and biological features can influence the data. It has been suggested that Sebutape, due to water occlusion and temperature insulation during the sampling period, interferes with sebum droplet formation and spreading. However, such systematic interference may be advantageous as sweating and heat are important clinical prerequisites in the formation of oiliness. Thus, it is believed that the Sebutape is a specialized method for the determination of 'oiliness', the very last phase of sebum output, in which sebum droplets spread over the skin surface.

The efficacy of this lotion is based on the synergistic action of ethyl linoleate and triethyl citrate, which are metabolized into active ingredients by *P. acnes*. Ethyl linoleate is hydrolysed to linoleic acid. It is known that linoleic acid is significantly reduced in epidermal and comedonal lipids of patients with acne, with subsequent reduction in the barrier functions of the epithelium, hyperkeratinization and conditions favouring bacterial proliferation. Linoleic acid levels return to normal with resolution of acne.^{10,11} Linoleic acid also downregulates neutrophilic oxygen metabolism and phagocytosis, and its low levels in epidermal lipids of patients with acne could contribute to the inflammation mediated by the neutrophils and in the oxidative damage *in situ* observed in acne lesions.¹² In addition, linoleic acid is a potent 5 α -reductase inhibitor and the low level of linoleic acid in acne-prone sebaceous glands could at least partly explain why these glands have abnormally high 5 α -reductase activity and therefore high sebum production.¹³ The enzymatic hydrolysis of ethyl linoleate in the new lotion produces linoleic acid resulting in reduction of seborrhoea due to its effect on 5 α -reductase, prevention of hypercornification and suppression of inflammation.

Triethyl citrate, the other component of this product, is hydrolysed by *P. acnes* to the diethyl ester and monoethyl ester of citric acid and to citric acid itself. As a result, the micro-environment of the pilosebaceous units becomes acidic, resulting in reduced proliferation of *P. acnes*, inhibition of bacterial lipases and 5 α -reductase and modulation of the keratolytic action exerted *in situ* by the carboxyl groups released in sequence by hydrolysis of the citric acid esters.

Therefore the combination of ethyl linoleate and triethyl citrate can reduce the hyperkeratinization of the pilosebaceous duct, the bacterial colonization of the infundibulum by *P. acnes* and seborrhoea, targeting the different steps in the pathogenesis of acne.

Present acne treatments have several shortcomings. Topical preparations are often irritating, cosmetically unacceptable and can bleach clothing or hair if they contain benzoyl peroxide. Oral antibiotics are effective in acne, but responses to treatment are typically slow and continuous treatment for 6–8 months is usually needed. This long-term exposure to antibiotics is causing a major concern due to the increase of antibiotic-resistant strains of *P. acnes* and, more significantly, *Staphylococcus epidermidis*, noted in patients treated for acne. Resistance in *P. acnes* may result in treatment failure. Resistance

in *S. epidermidis* may result in plasmid-mediated transfer of drug resistance to *S. aureus*.

Oral isotretinoin, a synthetic retinoid, is the most effective acne treatment currently available and induces long-term remission in a proportion of patients. The indications for use of isotretinoin have recently broadened from nodulocystic acne to less severe forms of acne, including mild to moderate disease failing to respond to systemic antimicrobials or acne associated with severe psychological problems.¹⁴ However, isotretinoin is highly teratogenic and women must avoid pregnancy during treatment and for 1 month after treatment. It frequently produces significant mucocutaneous symptoms and, less frequently, systemic symptoms such as myalgia, headaches and occasionally depression.^{15,16}

The ideal acne treatment should quickly and effectively control acne, have few if any side-effects, reduce seborrhoea and should be acceptable to patients. The patients in this trial are likely to have been broadly representative of adults with acne in the general population, although recruitment of volunteers might have introduced a selection bias towards those with long-standing acne that had failed conventional treatments. In addition, there was a 17.5% withdrawal from both groups, which is similar to the percentages noted in similar studies.

Our results demonstrate that this new topical lotion is an effective treatment for acne, with a rapid onset of action and observable improvement of acne within 4 weeks. The lotion is nonantibiotic based and its use would reduce the risk of antibiotic resistance developing within the skin flora. The most remarkable finding of this study is that this topical lotion significantly reduces sebum production in the patients treated, by a clinically evident amount. In a previous study, topical erythromycin-zinc complex has been shown to reduce sebum production by 20%. However, this new lotion has stronger sebosuppressive activity, reducing sebum activity by a mean of 53% after a period of 12 weeks. Other drugs that have been shown to be effective in reducing sebum excretion are oral isotretinoin and oral antiandrogens. Our study suggests that the new lotion containing triethyl citrate and ethyl linoleate is a significant new treatment for acne vulgaris.

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