Clinical Study Report

Sponsor:	Almirall Hermal GmbH
Study No.:	**H 552 000-0902 / 290102BS**
EudraCT-No.:	2009-009948-23
Title:	A phase IIa single-center, randomized, controlled, observer-blind study to investigate the antimicrobial efficacy of topical formulations containing octenidine and prednicarbate in an „expanded flora test“ with healthy subjects
Study Preparation:	**Study preparations:** 1) Octenidine/Prednicarbate cream (0.05 % octenidine, 0.25 % prednicarbate) 2) Octenidine/Prednicarbate cream (0.1 % octenidine, 0.25 % prednicarbate) 3) Octenidine/Prednicarbate cream (0.25 % octenidine, 0.25 % prednicarbate) 4) Active ingredient-free vehicle to study preparations 1 - 3 **Control:** 1) Octenisept (0.1 % octenidine, 2 % phenoxy ethanol) 2) Two untreated test fields
Clinical Phase:	IIa
Objectives:	Testing whether topical formulations containing octenidine plus prednicarbate exert an in vivo antibacterial action on skin surface bacteria multiplied by occlusion using the "expanded flora test”
Description:	This single-center study was controlled and observer-blind with random assignment of the treatments to the test fields. The study was performed in 20 male or female subjects with healthy skin in the test area. There were no dropouts. All 20 subjects were included in the intention-to-treat (ITT) and the per-protocol (PP) analyses. All subjects received all treatments. Altogether seven test areas on the back were occluded for 24 hours. Then the study preparations and controls were applied. Two occluded areas were left untreated as negative controls. After another 24 hours of occlusion the superficial flora was extracted and the number of CFUs (colony forming units) of the skin surface bacteria was evaluated.
Principal Investigator:	[redacted] bioskin GmbH Burchardstrasse 17, 20095 Hamburg, Germany
Clinical Trial Manager:	[redacted] bioskin GmbH Burchardstrasse 17, 20095 Hamburg, Germany
Project Manager Sponsor:	[redacted] Almirall Hermal GmbH Scholtzstrasse 3, 21465 Reinbek, Germany
GCP Compliance:	The study was conducted in compliance with Good Clinical Practice including the archiving of essential documents.
Study Dates:	June 02 to June 10, 2009
Date of Report:	September 24, 2009

2. Synopsis

Name of Company: Almirall Hermal GmbH	Individual Study Table Referring to Part of the Dossier Volume: Page:	*(For National Authority Use Only)*
Name of Finished Product:		
Name of Active Ingredient: Octenidine plus Prednicarbate		

Title of Study:
A phase IIa single-center, randomized, controlled, observer-blind study to investigate the antimicrobial efficacy of topical formulations containing octenidine and prednicarbate in an „expanded flora test“ with healthy subjects

Investigator(s):
[redacted]

Study center(s):
bioskin GmbH, Hamburg, Germany

Publication (reference):
Not applicable to this study

Studied period (years):	Phase of development:
2009	IIa

Objectives:
Testing whether topical formulations containing octenidine plus prednicarbate exert an in vivo antibacterial action on skin surface bacteria multiplied by occlusion using the "expanded flora test”

Methodology:
Altogether seven test areas on the back were occluded for 24 hours. Then 50 µl of the study preparations and controls were applied once to the respective previously marked and 24 hours occluded fields on the back. Two occluded areas were left untreated as negative controls. After another 24 hours of occlusion the superficial flora was extracted and the number of CFUs (colony forming units) of the skin surface bacteria was evaluated.

Number of subjects (planned and analyzed):
Twenty male or female subjects were included in the study. There were no dropouts. All 20 subjects were included in the ITT (intention-to-treat) and the PP (per-protocol) analyses.

Diagnosis and main criteria for inclusion:
Subjects with healthy skin in the area of the test fields, aged 18 to 45 years

Test product(s), dose and mode of administration, batch number:
Study preparations:
1) Octenidine/Prednicarbate cream (0.05 % octenidine, 0.25 % prednicarbate), batch no.: 910KK03
2) Octenidine/Prednicarbate cream (0.1 % octenidine, 0.25 % prednicarbate), batch no.: 910KK03
3) Octenidine/Prednicarbate cream (0.25 % octenidine, 0.25 % prednicarbate), batch no.: 910KK03
4) Active ingredient-free vehicle to study preparations 1-3, batch no.: 910KK03
single topical occlusive application of approx. 50 µl formulation per test field (16.0 cm²)

Duration of treatment:
24 hours

Reference therapy or controls, dose and mode of administration, batch number:
Control:
1) Octenisept (0.1 % octenidine, 2 % phenoxy ethanol), batch no.: 908K0194
2) Two untreated test fields
single topical occlusive application of approx. 50 µl formulation per test field (16.0 cm²)

Duration of treatment:
24 hours

2. Synopsis (continued)

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Criteria for evaluation:

Primary efficacy variable: Colony forming unit counts for skin surface bacteria was the primary variable.

Safety: Screening and final clinical examinations, recording of adverse events.

Statistical Methods:

Analysis populations

Efficacy population

The Full Analysis Set included all randomized subjects who received at least one dose of study medication and had a valid assessment of the primary parameter. The intention-to-treat (ITT) analysis was based on the Full Analysis Set.

The Valid-Cases Set included all subjects in the Full-Analysis Set, excluding subjects with major protocol violations or significant protocol deviations.

Major protocol violations included but were not limited to:

- inappropriate enrollment,
- the use of prohibited concomitant medication,
- reaching a major exclusion criterion during the trial.

Significant protocol deviations included:

- identified protocol violations or significant deviations during the “Subject Data Inclusion” meeting.

The per-protocol (PP) analysis was based on the Valid-Cases Set.

Safety population

The Safety Set included all randomized subjects who received at least one application of study medication. All safety analyses were based on the Safety Set.

Efficacy analyses

Hypotheses

No formal hypotheses were postulated. All inferential statistics were interpreted exploratory.

Statistical analyses

The **number of bacteria per cm² skin surface** was determined as:

$$\frac{K \times 20}{V \times 3.83\ cm^2}$$ K = number of colonies and V = dilution

With the dilution factor (V) as 1 for the undiluted solution and 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5} for the dilutions 1:10, 1:100, 1:1,000, 1:10,000 and 1:100,000.

2. Synopsis (continued)

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Statistical Methods (continued):

The number of bacteria calculated for the test areas was given in relation to the average number of germs from the two negative control fields. Since bacteria numbers could be considered to have a logarithmic normal distribution, the bacteria numbers were expressed as logs.

The primary parameter **Log untreated control corrected number of bacteria per skin surface [log count/cm²]** was then calculated as:

Z = log (average of the bacteria number from the control fields + 1)
– log (number of bacteria from test area + 1)

The primary variable Log untreated control corrected number of bacteria per skin surface [log count/cm²] and number of bacteria per cm² skin surface were summarized by descriptive statistics (number of valid values, mean, standard deviation, median, interquartile range, minimum and maximum) for each treatment.

The study preparations were compared with respect to the mean of the primary parameter. Descriptive hypothesis test was performed testing the implicit null hypothesis

H_{01}: The two study preparations did not differ in their mean effectiveness

against the alternate hypothesis:

H_{11}: The two study preparations differed in their mean effectiveness.

The two-sided hypotheses were tested applying the paired t-test at a significance level of 5 %. The following comparisons were performed:

1.	Octenidine/Prednicarbate cream (0.25 % octenidine, 0.25 % prednicarbate)	vs.	Active ingredient-free vehicle
2.	Octenidine/Prednicarbate cream (0.1 % octenidine, 0.25 % prednicarbate)	vs.	Active ingredient-free vehicle
3.	Octenidine/Prednicarbate cream (0.05 % octenidine, 0.25 % prednicarbate)	vs.	Active ingredient-free vehicle
4.	Octenidine/Prednicarbate cream (0.25 % octenidine, 0.25 % prednicarbate)	vs.	Octenidine/Prednicarbate cream (0.05 % octenidine, 0.25 % prednicarbate)
5.	Octenidine/Prednicarbate cream (0.25 % octenidine, 0.25 % prednicarbate)	vs.	Octenidine/Prednicarbate cream (0.1 % octenidine, 0.25 % prednicarbate)
6.	Octenidine/Prednicarbate cream (0.1 % octenidine, 0.25 % prednicarbate)	vs.	Octenidine/Prednicarbate cream (0.05 % octenidine, 0.25 % prednicarbate)

2. Synopsis (continued)

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Statistical Methods (continued):

Statistical analyses (continued)

Following the closed set procedure, all significant comparisons up to the first not significant comparison preserved the family wise type I error rate of 5 %. The following comparisons were reported for descriptive reasons only.

Additionally Octenisept® (0.1 % octenidine, 2 % phenoxy ethanol) was tested vs. active ingredient-free vehicle according to the upper hypothesis. The comparison was interpreted descriptively.

Safety analyses

Adverse events: Tables with adverse events are presented as appropriate.

Summary, conclusions:

Efficacy results:

Under the conditions in this “expanded flora test” with modified occlusion times all three active study preparations containing octenidine plus prednicarbate exerted an in vivo antibacterial action on skin surface bacteria.

An increasing antibacterial action was detected depending on the concentration of octenidine in the prednicarbate/octenidne-combination.

The greatest antimicrobial effect was noted for the combination Octenidine 0.25 %/Prednicarbate 0.25 % cream (mean Z = 4.55). Stepwise lower reductions of the number of CFUs (colony forming units) of skin surface bacteria were noted for the combination Octenidine 0.1 %/Prednicarbate 0.25 % cream (mean Z = 3.07) and Octenidine 0.05 %/Prednicarbate 0.25 % cream (mean Z = 1.97). A comparable antimicrobial effect to the Octenidine 0.1 %/Prednicarbate 0.25 % cream was noted with the marketed product Octenisept® (mean Z = 2.71)), which both have an octenidine concentration of 0.1%.

No relevant effect on skin surface bacteria was noted for the active ingredient-free vehicle (mean Z = -0.03).

In the statistical comparisons to the active ingredient-free vehicle, all four active formulations showed significantly greater mean Log untreated control corrected numbers of bacteria per skin surface (mean Z-values) ($p < 0.0001$, respectively).

Moreover, in the statistical comparison between the three investigational products, a significantly greater mean Log untreated control corrected number of bacteria per skin surface (mean Z-values) was noted for the combination Octenidine 0.25 %/Prednicarbate 0.25 % cream when comparing to the other two formulations Octenidine 0.05 %/Prednicarbate 0.25 % cream ($p < 0.0001$) and Octenidine 0.1 %/Prednicarbate 0.25 % cream ($p < 0.0004$). In addition, the combination Octenidine 0.1 %/Prednicarbate 0.25 % cream showed a significantly greater mean Log untreated control corrected number of bacteria per skin surface (mean Z-values) than Octenidine 0.05 %/Prednicarbate 0.25 % cream ($p = 0.0069$).

Safety results:

Altogether, 11 non-serious adverse events were reported in three subjects in this study.

Three AEs in one subject were identified as contact dermatitis and classified as moderate and definitely related to study medication. These local AEs occurred one day after study end and had to be treated with Ecural® Salbe. A follow-up twelve days after study end revealed that erythema and itching were no longer present and the test fields showed only mild hyperpigmentation.

2. Synopsis (continued)

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Summary, conclusions:

Safety results (continued)

Seven local AEs in another subject were identified as irritant contact dermatitis on the last study day and classified as mild and were considered to be unlikely related to study medication. A follow-up one day later revealed an improvement of the affected test fields, but the test fields were nonetheless treated once with Ecural® Salbe.

One mild AE in a further subject was unlikely related to the study medication and the subject recovered without sequelae.

The final physical examination did not show relevant findings in any of the subjects and there were no other relevant observations related to safety in this study.

Conclusion:

The aim of the study was to investigate the antimicrobial efficacy of topical formulations containing octenidine and prednicarbate in an “expanded flora test” in healthy subjects.

The three study preparations (Octenidine 0.05 %/Prednicarbate 0.25 % cream, Octenidine 0.1 %/Prednicarbate 0.25 % cream and Octenidine 0.25 %/Prednicarbate 0.25 % cream) exerted a clear, significant in vivo antibacterial action on skin surface bacteria under the conditions in this study. In this “expanded flora test” a bacterial reduction of 2 to 4 magnitudes was found which reflects a clear antibacterial action according to literature (5).

The study preparation with the highest octenidine concentration, Octenidine 0.25 %/Prednicarbate 0.25 % cream, showed the greatest antimicrobial effect with a bacterial reduction of 4 magnitudes.

A somewhat lower antimicrobial effect (reduction of 3 to 2 magnitudes) was noted for the two formulations Octenidine 0.1 %/Prednicarbate 0.25 % cream and Octenidine 0.05 %/Prednicarbate 0.25 % cream, whereas the reduction in skin surface bacteria of the Octenidine 0.1 %/Prednicarbate 0.25 % cream was comparable to the reducing effect seen for the control Octenisept®. Thus, the study preparations with different concentrations of octenidine revealed a clear positive dose-response antibacterial action with a bacterial reduction of 4 magnitudes for the highest tested concentration. The comparison of Octenidine 0.1 %/Prednicarbate 0.25 % cream with the marketed product Octenisept® ; which contains octenidine also in the concentration of 0.1 %, showed a clear antibacterial effect, which was in the same range for both products.

No relevant effect on skin surface bacteria was noted for the active ingredient-free vehicle. The number of skin surface bacteria was comparable to the untreated field (approximately 10^6, both). Thus, the number of skin surface bacteria after occlusion of the untreated control fields is comparable to the values described in the literature for the untreated control fields.

One subject developed a contact dermatitis in the test fields treated with all three study medications which was considered to be definitely related to study medications. The test fields had clearly improved at the follow-up visit twelve days later. There were no other safety concerns based on the results of this study.

Date of the report: September 24, 2009