



***In-vivo* Immediate and Long Term Effects**

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Tradename: AcquaSeal® Algae

Code: 20852

CAS #: N/A

Test Request Form #: 3607

Lot #: NC170831-I

Sponsor: Active Concepts, LLC; 107 Technology Drive Lincolnton, NC 28092

Study Director: Maureen Danaher

Principle Investigator: Jennifer Goodman

Test Performed:

In-vivo Immediate and Long Term Analysis

Purpose

To determine the ability of **AcquaSeal® Algae** when used at 2.0% in a simple lotion to modify the skin cosmetic properties *in vivo* after a single use and with repeated use. A single use fifteen (15) subject study evaluated the immediate impact of **AcquaSeal® Algae** on skin properties. A fifteen (15) subject study over a four week period evaluated the longer term and cumulative effects of **AcquaSeal® Algae** on skin properties related to cosmetic benefits and skin health.

Materials & Methods

Single Use Study

A single use fifteen (15) subject study evaluated the immediate impact of **AcquaSeal® Algae** on skin properties. **AcquaSeal® Algae** was formulated into a lotion base at 2.0% and applied to the face (one side) of test panelists. To the other side the control lotion was applied. A number of skin parameters were assessed after one hour. Measurements included skin feel, skin friction and skin hydration. Tests were run in the morning and subjects refrained from using any cosmetic product on their faces for at least three days prior to the test. Subjects equilibrated in a controlled environment room for thirty minutes prior to the test.

Skin hydration was assessed with the DPM value of the Nova Impedance Meter as per manufacturer's instructions. Measurements were made in triplicate and averaged. Skin slip and overall feel was self-assessed on a 0-10 Clinical Scale, subjects being supervised by trained clinicians. Subjects would place a line of card on which a visual linear 0-10 scale had been placed.

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Friction was assessed with a Courage Friction Meter FR 700 as per manual instructions for measuring skin friction using a 16mm head. Data was acquired as AFU and converted with proprietary software to a 1-10 scale wherein 1 was a synthetic skin treated with dimethicone to dramatically reduce slip or friction and 10 was the skin mimic treated with a sucrose/ water solution to maximize resistance. Measurements are essentially arbitrary with respect to specific numbers and all results should be viewed comparatively, i.e. A versus B or before versus after. Skin-feel and skin slip were graded on a 1-10 clinical scale with subjective self-assessment under supervision of trained clinicians.

Long Term (Four Week) Clinical Study

A fifteen (15) subject study over a four week period evaluated the longer term and cumulative effects of **AcquaSeal® Algae** on skin properties related to cosmetic benefits and skin health. After an initial one week dry out phase (no cosmetic product use) subjects (average age 52) applied a 2.0% **AcquaSeal® Algae** lotion to their half of their face twice-a-day for four weeks. The placebo was applied to the other half. Evaluations were made prior to the study start and after four weeks. At the four week evaluation, assessments were made at least six hours after production application, i.e. applications were made first thing in the morning and prior to retiring, thus measurements were taken mid to late afternoon following the previous morning application.

Hydration was assessed with the NOVA meter (DPM value) as per manufacturer’s instructions. All measurements were done in triplicate. Superficial facial lines (SFL’s) were assessed (half face) by trained graders using the method of Packman (Packman, E., and Gans, E.H. Topical moisturizers: quantification of the effect on superficial facial lines. Soc. Cos. Chem. 29, 79-90 (1992). In this method a total wrinkle or SFL is determined based upon the summation of the number of wrinkles, fine (1), moderate, (2) and deep (3). The number in parenthesis refers to a multiple weight factor for each wrinkle class. Clinical grading of overall skin condition was done via expert graders on a 0-10 point scale as above using a visual linear 0-10 scale.

Clarity (luminosity L value) was assessed with the Minolta Chroma Meter (Muizzuddin, N., Marenus, K., Maes, D., and Smith, W. Use of a chromameter in assessing the efficacy of antiirritants and tanning accelerators. J. Soc. Cos. Chem. 1990.).

Skin Firmness was assessed with a Cutometer 575 using the ratio of Ue/Ur as a measure of firmness.

Placebo Composition Formulation

Phase A

Deionized Water	62.600%
Magnesium Aluminum Silicate	0.400%
Xanthan Gum	0.150%
Acrylates/C10-30 AlkylAcrylate Crosspolymer	0.750%

Phase B

Butylene Glycol	4.000%
Disodium EDTA	0.050%

Phase C

Caprylic/Capric Triglyceride	8.500%
Octyl Palmitate	4.000%
Cetearyl Alcohol	2.000%

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PEG-8 Stearate	1.000%
PEG-100 Stearate	0.800%

Phase

Triethanolamine 99%	0.100%
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Phase E

Water	14.000%
Phenoxyethanol	0.500%
Potassium Sorbate	0.100%
Methylisothiazolinone	0.050%
Butylene glycol	1.000%

Results

Single Use Study

As shown in Figure 1 below, the placebo did not significantly alter skin hydration or feel or hydration characteristics after one hour. **AcquaSeal® Algae** on the other hand, significantly improved skin hydration, reduced skin friction and provided a positive skin feel.

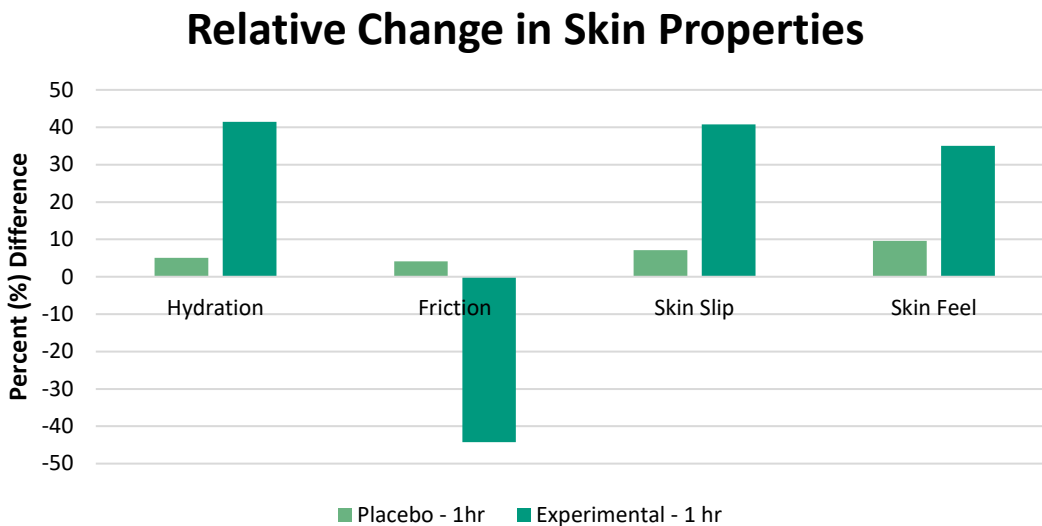


Figure 1. Relative change in skin properties after one hour.

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Long-Term Use-Long Term Skin Changes

Summarized below in Figure 2 are the results after one month product usage; results are expressed as percent change from the pre-treatment assessments, both for the clinical and laboratory evaluations. **AcquaSeal® Algae** produced considerable positive changes in skin quality after both four weeks. Statistically significant improvements in skin hydration, clarity, lines and wrinkles, firmness, flakiness and overall appearance were observed with continuing use of **AcquaSeal® Algae**. The placebo showed no such changes.

Relative Change in Skin Properties

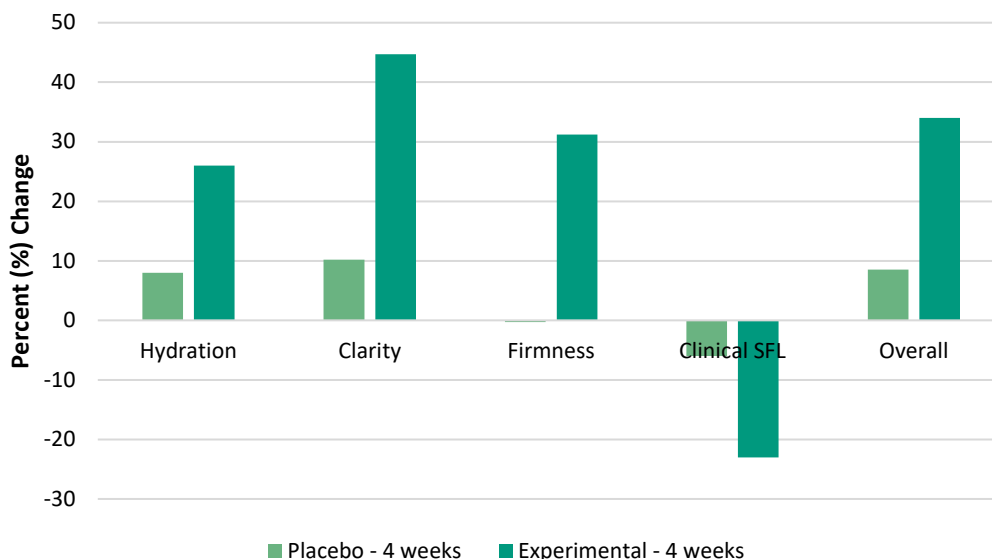


Figure 2. Percent change in skin properties after four weeks.

Discussion

Based on the results in these studies, **AcquaSeal® Algae** is capable of exhibiting both short term and long term skin benefits. Short term use of 2.0% **AcquaSeal® Algae** in a base lotion showed a 41.4% increase in hydration, 44.2% decrease in friction, 40.7% increase in skin slip, and 35.0% increase in skin feel. Long term use of 2.0% **AcquaSeal® Algae** in a base lotion over four weeks showed a 26.0% increase in hydration, 44.7% increase in clarity, and 31.2% increase in firmness. These results support using **AcquaSeal® Algae** in formulations for both short term and long term cosmetic benefits.