AcquaSeal® Coconut Efficacy Data

Code: 20742
INCI Name: Cocos Nucifera (Coconut) Fruit Extract
CAS #: 8001-31-8
EINECS #: 232-282-8

Name of Study	Results
24 Hour Moisturization Assay	An *in-vivo* study was conducted on 10 (M/F) subjects over a period of 24 hours to evaluate the moisturization benefits of **AcqualSeal® Coconut**. Results indicate that this material is capable of significantly increasing moisturization on the skin after 24 hours than the control.
Coefficient of Permeability	The purpose of this study is to determine the relative permeability coefficients of several materials used for the development of barrier layers on the skin. A higher number for the Coefficient of Permeability means that more liquid was able to seep through into the solution, yielding lower barrier function. **AcquaSeal® Coconut** has a low Coefficient of Permeability providing better barrier function than most alternatives, as it does not allow for moisture loss.
Hydration Potential	This study will demonstrate the hydration potential of natural, synthetic and animal derived materials for comparison via their respective water holding capacities. The results indicate that **AcquaSeal® Coconut** is an effective all-natural and botanical alternative to Lanolin as it is capable of holding 200% of its weight in water.
Lipophilic ORAC Assay	This assay was conducted to assess the antioxidant capacity of **AcquaSeal® Coconut**. **AcquaSeal® Coconut** exhibited antioxidant activity comparable to the known standard antioxidant, Trolox®. The antioxidant capacity of **AcquaSeal® Coconut** increased

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	as the concentration increased, as a result we can assure that its ability to minimize oxidative stress is dose dependent.

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AcquaSeal® Coconut 24 Hour Moisturization Study

Code: 20742
INCI Name: Cocos Nucifera (Coconut) Fruit Extract
Text Request Number:
Suggested Use Levels: 0.50 - 5.00%

Abstract

An in-vivo study was conducted over a period of 24 hours to evaluate the moisturization benefits of AcqualSeal® Coconut. Ten (M/F) subjects between the ages of 23-45 participated in the study. Results indicate that this material is capable of significantly increasing moisturization on the skin after 24 hours than the control.

Materials and Methods

Ten volunteers M/F between the ages of 23 and 45, known to be free of any skin pathologies participated in this study. A DermaLab Corneometer was used to measure the moisture levels on the subject's volar forearms. The Corneometer is an instrument that measures the amount of water within the skin. The presence of moisture in the skin improves conductance, therefore results in higher readings than dry skin. Hence, the higher the levels of moisture, the higher the readings yielded from the Corneometer. Baseline moisturization readings were taken when the study commenced (t=0).

Following initial measurements, all subjects were asked to apply 2 mg of each test material to their volar forearms. Measurements were taken immediately after application of the test materials and then again 24 hours after application of the test materials. The experimental material consisted of 2.0% AcquaSeal® Coconut in a base lotion.

For added perspective, measurements of an untreated test site and a site treated with a base lotion (Cetaphil Moisturizing for All Skin Types) were recorded.

Results

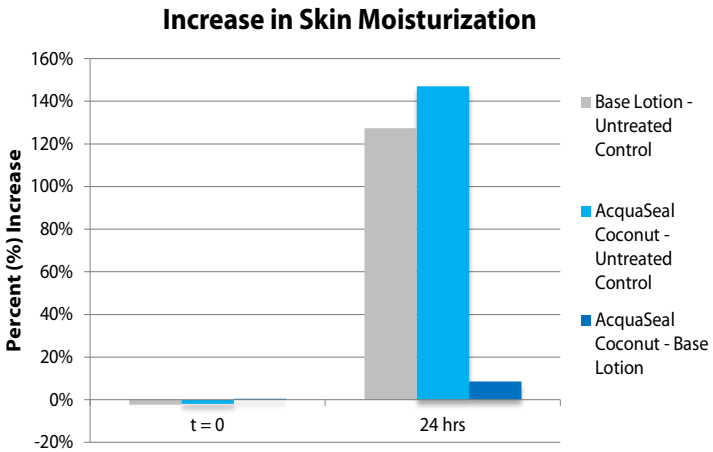

Figure 1. Percent increase in moisturization 24 hours after application of AcquaSeal® Coconut and base lotion

Discussion

As evidenced in the 24 hour efficacy study of AcquaSeal® Coconut on the skin, moisture levels were improved by 150% after 24 hours when compared to the untreated control. When compared to the base lotion, AcquaSeal® Coconut improved moisturization by 14% after 24 hour hours. Results indicate that AcquaSeal® Coconut is capable of increasing moisturization more effectively than the base lotion after 24 hours.

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AcquaSeal® Coconut Coefficient of Permeability

Code: 20742
INCI Name: Cocos Nucifera (Coconut) Fruit Extract
Text Request Number: 210
Suggested Use Levels: 0.50 - 5.00%

Abstract

The purpose of this study is to determine the relative permeability coefficients of several materials used for the development of barrier layers on the skin. The coefficient of permeability is determined by a gravimetric analysis of moisture transfer across a semi-occluded membrane.

Materials and Methods

The sample oil and Mineral Oil 70 were mixed together in a 1:1 ratio. The mixture was applied to filter, which was then placed on top of a measurement cup containing $CaCl_2$ solution. These were allowed to stand for 24 hours at 25^0C with 95% RH, and the weight of the moisture that permeated through the filter paper and into the solution was measured as increased weight.

The Coefficient of Permeability was shown in a percentage through comparing an increase in weight to a control where no oils were applied.

Results

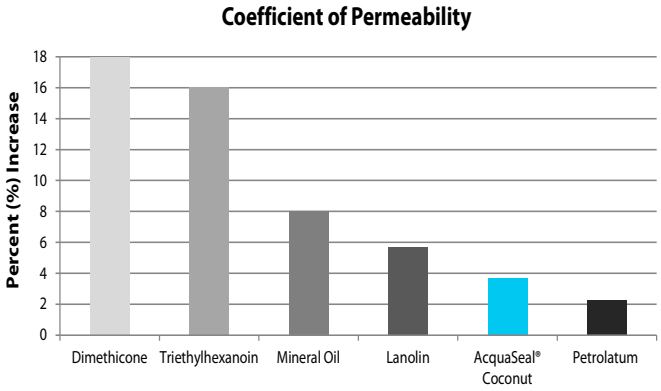

Figure 1. Coefficient of Permeability Measurements

Discussion

A higher number for the Coefficient of Permeability means that more liquid was able to seep through into the solution, and thus a lower barrier function. According to Figure 1., **AcquaSeal® Coconut** has a lower Coefficient of Permeability than Dimethicone, Triethylhexanoin, Mineral Oil, and Lanolin, which means that AcquaSeal® Coconut provides better barrier function than all of these products, as it does not allow for moisture loss.

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Hydration Potential

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Tradename: AcquaSeal® Coconut

Code: 20742

CAS #: 8001-31-8

Test Request Form #: 211

Lot #: 57669P

Sponsor: Active Concepts, LLC; 107 Technology Drive Lincolnton, NC 28092
Study Director: *Erica Segura*
Principle Investigator: *Maureen Danaher*

Abstract

The purpose of this study is to determine the relative hydration potential of several materials which exhibit moisturizing properties through their respective water hold capacities. This study will demonstrate the hydration potential of 2.0% **AcquaSeal® Coconut** in comparison to some of its natural, synthetic, and animal-derived competitors.

Materials and Methods

Hydration Potential was measured according to the British Pharmacopoeia (BP) water absorption capacity method. Following this procedure, sample materials were placed separately into a mortar. Water was then incrementally added to the sample and mixed using a pestle. Samples, each tested at 2.0%, were considered to be saturated when no more water can be mixed into the emulsion. The point at which a sample is fully saturated is referred to as the terminal point. The water holding capacity was then calculated by dividing the weight of the sample after the terminal point has be reached by the initial sample weight and multiplying by 100 as is indicated in the below equation.

Water Holding Capacity (%) = (Weight of sample after terminal point is reached/Weight of Initial Sample) x 100

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Results

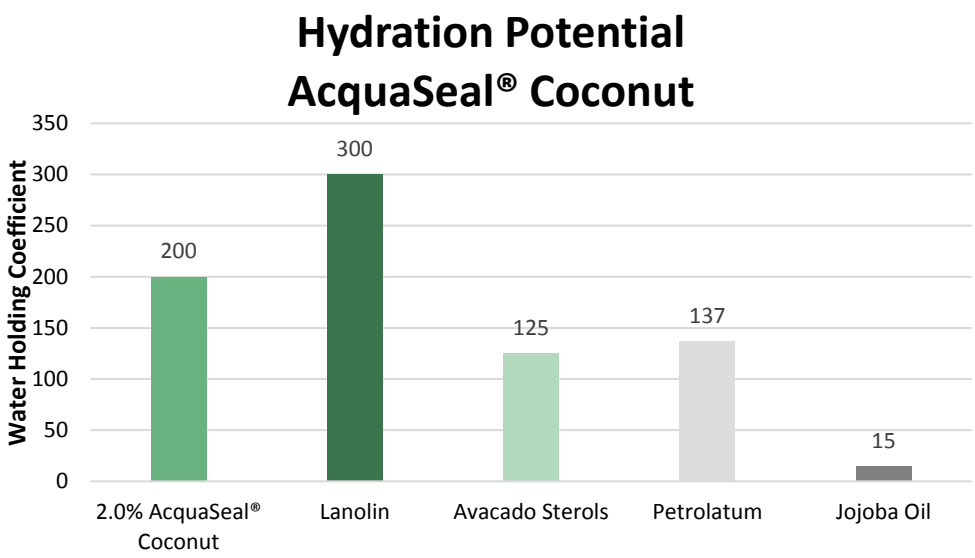

Figure 1. Hydration Potential Results

Discussion

The results indicate that **AcquaSeal® Coconut** is an excellent all natural and botanical alternative to Lanolin as it is capable of holding 200% of its weight in water. Data analysis also reveals that compared to Avocado Sterols, Petrolatum, and Jojoba Oil, **AcquaSeal® Coconut** exhibited superior hydration potential with respective improvements in water holding capacity of 46%, 37% and 172%, respectively. These findings confirm that **AcquaSeal® Coconut** is useful in topical applications to effectively deliver moisture to the skin.

Oxygen Radical Absorbance Capacity (ORAC) Assay

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Tradename: AcquaSeal® Coconut

Code: 20742

CAS #: 8001-31-8

Test Request Form #: 58

Lot #: NC120625-A

Sponsor: *Active Concepts, LLC; 107 Technology Drive Lincolnton, NC 28092*
Study Director: *Erica Segura*
Principle Investigator: *Meghan Darley*

Test Performed:
Oxygen Radical Absorbance Capacity (ORAC)

Introduction

Reactive oxygen species (ROS) are generated by normal cellular processes, environmental stresses, and UV irradiation. ROS are dangerous to cellular structures and functional molecules (i.e DNA, proteins, lipids) as they act as strong oxidizing agents or free radicals. The oxygen radical absorbance capacity (ORAC) assay is a standard method used to assess antioxidant capacity of physiological fluids, foods, beverages, and natural products. The assay quantitatively measures a sample's ability to quench free radicals that have the potential to react with and damage cellular components.

Oxygen Radical Absorbance Capacity (ORAC) assay was conducted to assess the antioxidant capacity of **AcquaSeal® Coconut**.

Assay Principle

This assay is based upon the effect of peroxyl radicals generated from the thermal decomposition of 2, 2'-azobis-2-methyl-propanimidamide dihydrochloride (AAPH) on the signal intensity from the fluorescent probe, fluorescein, in the presence of an oxygen radical absorbing substance. The degree of change is indicative of the amount of radical damage and the presence of antioxidants results in an inhibition in the free radical damage to the fluorescein. The antioxidant protection of the sample can be calculated by comparing it to a set of known standards. Trolox®, a water soluble vitamin E analog, with known antioxidant capabilities is used in this ORAC assay as the standard for measuring the antioxidant capacity of unknown substances. ORAC values, expressed in µM of Trolox® equivalents (TE), are calculated using the area under the curves (AUC) of the test product, Trolox®, and the control materials. Trolox equivalency is used as the benchmark for antioxidant capacity of mixtures since it is difficult to measure individual components.

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Materials

A. Equipment:	Synergy H1 Microplate reader (BioTek Instuments, Winooski, VT); Gen5 software (BioTek Instuments, Winooski, VT); Pipettes
B. Buffers:	75mM Potassium Phosphate (pH 7.4); Deionized H_2O; Acetone
C. Reagents:	2,2′-Azobis(2-methylpropionamidine) dihydrochloride (AAPH) (153mM); 6-Hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox®); Fluorescein Sodium Salt (4nM)
D. Preparation:	Pre-heat (37°C) Synergy H1 Microplate reader; Prepare Trolox® standards, sample dilutions, fluorescein solution, and AAPH.
E. Microtitre Plates:	Corning 96 Well Black Side/Clear Bottom Microplates

Methods

Solutions of **AcquaSeal® Coconut** and Trolox® (positive control) were prepared in a 1:1 Acetone:DiH_2O. Materials were prepared at three different concentrations/dilutions. Trolox® was used as a reference for antioxidant capacity and prepared at a concentrations ranging from 12.5µM to 200µM in a 1:1 Acetone:DiH_2O.

For the ORAC assay, 25µL of test material and Trolox® were combined with 150µL of fluorescein in 75mM potassium phosphate buffer and incubated in the Synergy HT Microplate reader at 37˚C for 30 minutes. At the end of the incubation period, 25µL of AAPH were pipetted into each well. Fluorescent measurements were then taken every 2 minutes for approximately 2 hours at an excitation wavelength of 485nm and an emissions wavelength of 520nm.

The AUC and Net AUC values of the standards and samples were determined using Gen5 2.0 Data Reduction Software using the below equations:

$$AUC = 0.5 + \frac{R2}{R1} + \frac{R3}{R1} + \frac{R4}{R1} + \cdots + \frac{Rn}{R1} \rightarrow Where\ R\ is\ fluorescence\ reading$$

$$Net\ AUC = AUC_{sample} - AUC_{blank}$$

The standard curve was obtained by plotting the Net AUC of different Trolox® concentrations against their concentration. ORAC values of samples were then calculated automatically using the Gen5 software to interpolate the sample's Net AUC values against the Trolox® standard curve. ORAC measurements for the test material were expressed in micro moles Trolox® equivalents (µMTE), where 1 ORAC unit is equal to 1 µMTE.

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Results

AcquaSeal® Coconut began exhibiting antioxidant activity at a 0.05% concentration.

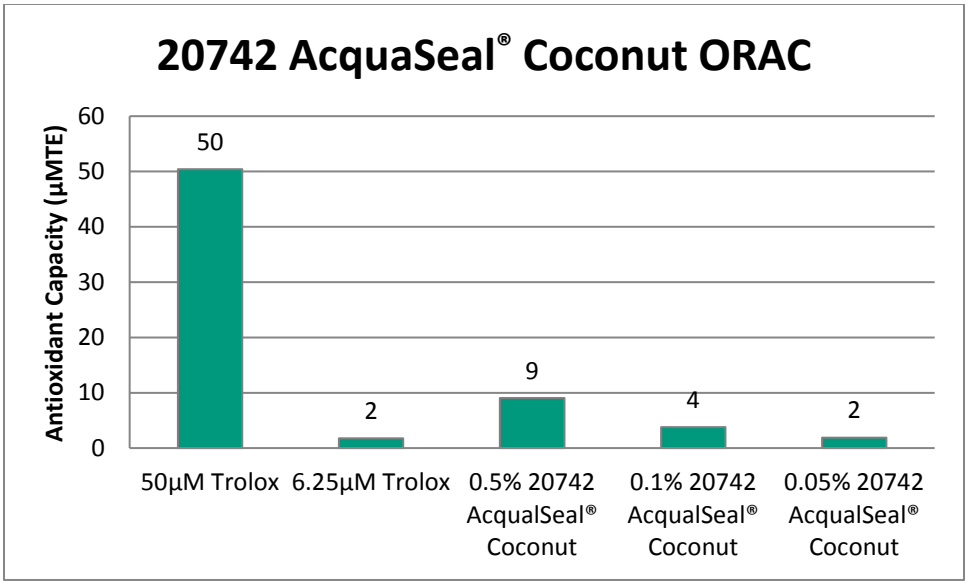

Figure 1: Antioxidant capacities

Discussion

As shown in figure 1, **AcquaSeal® Coconut** exhibited antioxidant activity comparable to out known standard antioxidant, Trolox®. The antioxidant capacity of **AcquaSeal® Coconut** increased as the concentration increased, as a result we can assure that its ability to minimize oxidative stress is dose dependent.

AcquaSeal® Coconut was designed to be moisturizing, nourishing, and hydrating, and provide anti-aging and relipidizing properties. With the present study we can confirm that this unique ingredient is not only capable of providing functional benefits but it is also capable of providing potent antioxidant benefits when added to cosmetic applications.