

**Tradename:** Phytofuse Renew®

**Code:** 16586

**CAS #:** 90106-73-3

**Test Request Form #:** 3974

**Lot #:** NC180213-C

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

**Study Director:** *Erica Segura*

**Principal Investigator:** *Maureen Danaher*

**Test Performed:**

Carbon Pollution Protection Study

**Introduction**

The role of pollution in the appearance of premature wrinkles and age spots has become a new frontier in antiaging active ingredients. While we have known about the harmful effects of pollution on our health for years, new research indicates air pollution plays a detrimental role in extrinsic aging. Carbon and metal micro particles found in polluted air embedded in the dermis cause oxidative stress, initiate inflammatory cascade leading to the breakdown of collagen, elastin, and other structural components in the skin. Additionally, polyaromatic hydrocarbons overstimulate the aryl hydrocarbon receptors on keratinocytes and melanocytes resulting in hyperpigmentation and the appearance of age spots. Providing a physical barrier will prevent embedment of carbon particles, thus reducing the signs of extrinsic aging.

Accordingly, a Carbon Pollution Protection Study was conducted to assess the ability of **Phytofuse Renew®** to provide immediate barrier protection from carbon air pollution and enhance the removal of carbon air pollution.

**Study Principle**

Products are applied to the skin and micronized charcoal is applied on top. The micronized charcoal used has a particle size of 2.5 microns (PM 2.5) or less that mimics the small particulates found in polluted air. Pictures of the charcoal are analyzed for the amount of carbon on the skin before and after a washing procedure to determine the amount of carbon present on the skin.

**Materials**

- A. Equipment:** DermaLab Skin Combo (DermaScope Camera Probe); ImageJ Analysis Software (NIH); Pipettes
- B. Reagents:** Base Cleanser (Cetaphil® Cleanser for All Skin Types); Micronized activated charcoal;  
Deionized water
- C. Other:** Disposable pipette tips; Wash bottles
- D. Software:** Excel Analysis ToolPak (Microsoft)

## Methods

Ten volunteers, male and female, between the ages of 23 and 45, who were known to be free of any skin pathologies with Fitzpatrick skin types I to III, participated in this study (Table 1).

**Table 1.** The Fitzpatrick Classification of Skin Types Chart<sup>1</sup>

Fitzpatrick Skin Type Descriptions*	
Skin Type	Description
I	Always burns, never tans
II	Burns easily, tans minimally
III	Burns moderately, tans to light brown
IV	Burns minimally, tans to moderate brown
V	Rarely burns, tans to dark
VI	Never burns, least sensitive to changes

\*Adapted from The Surgeon General's Call to Action to Prevent Skin Cancer

Three randomly designated test sites were identified on the volar forearm and assigned a condition (Table 2). The skin test site conditions and treatments are described below (Table 2). The Base Cleanser utilized in this study was Cetaphil® Cleanser for All Skin Types. After participants applied 0.2 g of each treatment to the assigned test site, the Cleansers were allowed to dry completely. Participants applied 5 mg of micronized charcoal on top of each test site and initial images were obtained. After each test site was washed 5 times with deionized water, images were obtained again.

**Table 2.** Descriptions of the Conditions and Treatments for each Skin Test Site

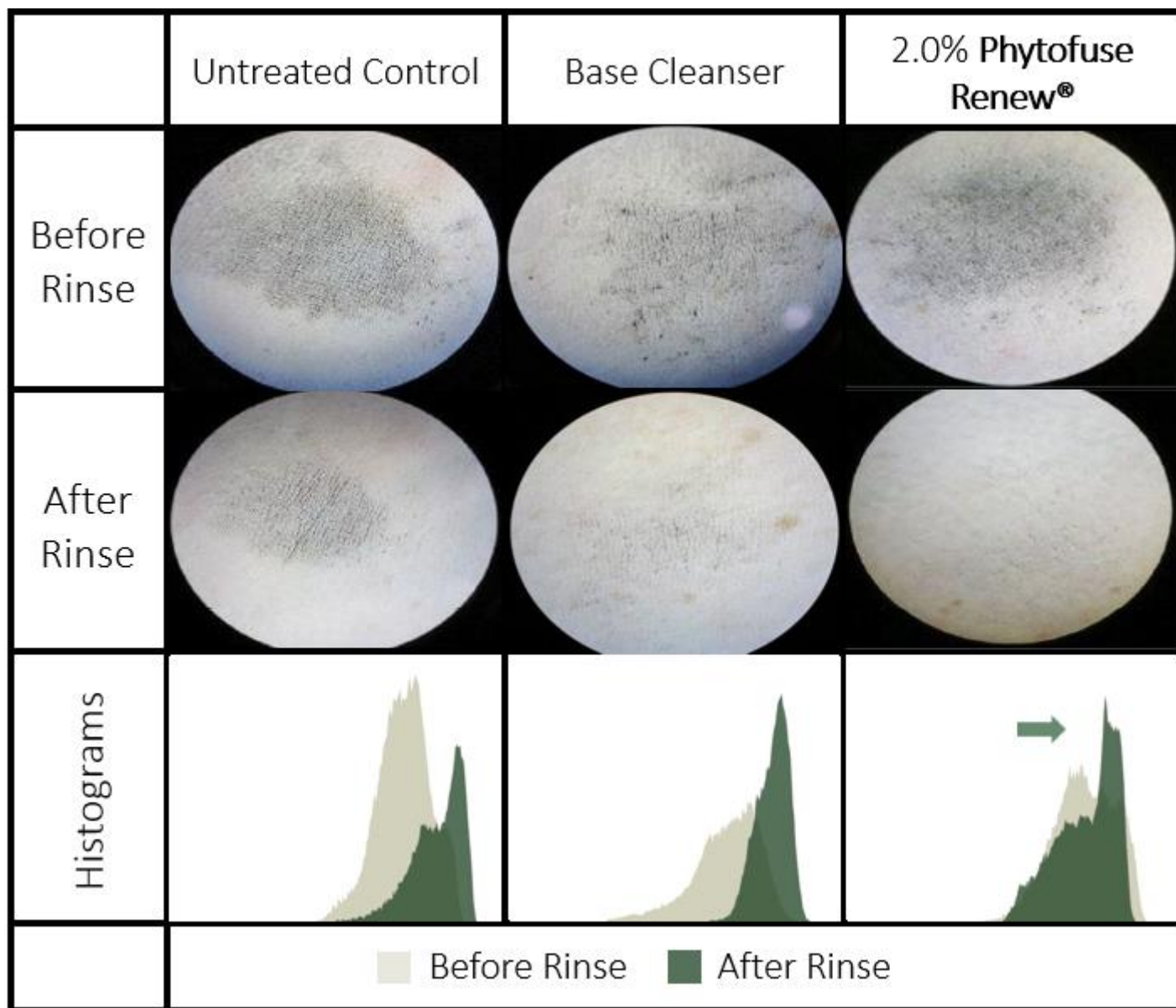
Skin Test Site	Condition	Treatment / Test Article Application Description
1	Untreated Control	None
2	Base Cleanser	Base Cleanser
3	2.0% <b>Phytofuse Renew</b> ®	2.0% <b>Phytofuse Renew</b> ® in Base Cleanser

ImageJ was utilized to determine color intensity from the captured images. The histogram results are presented as shifts in light intensity. The left side of the histograms indicate more dark light intensity, whereas the right side of the histograms indicate more lighter intensity. Data was analyzed using a one-way ANOVA with statistical significance accepted at  $p \leq 0.05$ . The percent change in histogram light intensity was calculated for each test site using the following equation:

$$\text{Percent Change (\%)} = \frac{\text{Light Intensity}_{\text{After Wash}} - \text{Light Intensity}_{\text{Before Wash}}}{\text{Light Intensity}_{\text{Before Wash}}} \times 100$$

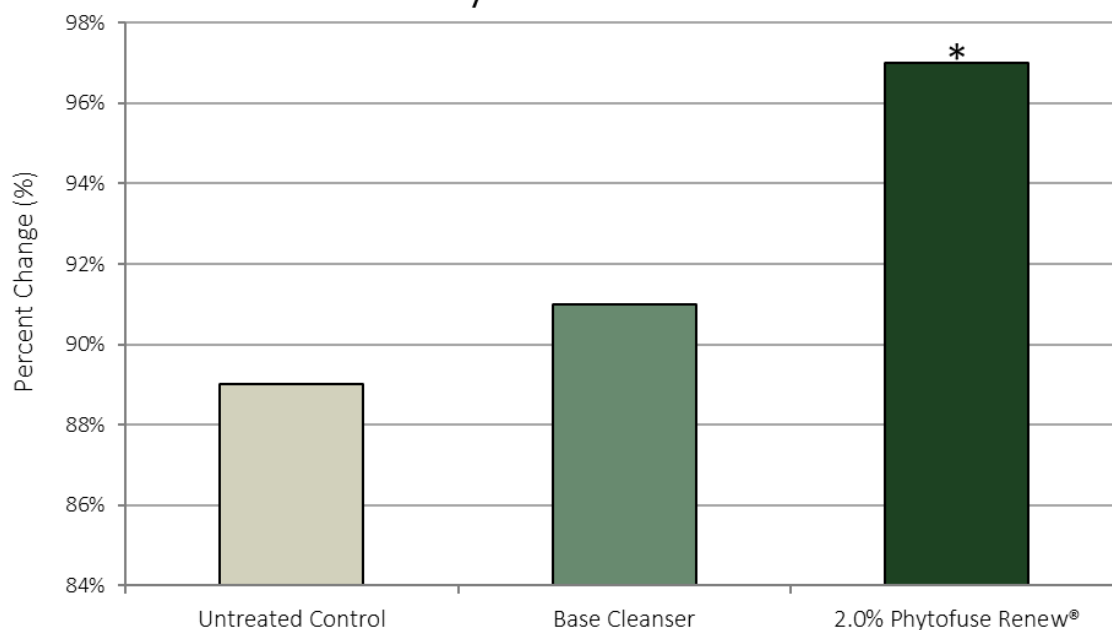
## Results

The data obtained from this study met criteria for a valid study as the Untreated Control and Base Cleanser performed as anticipated. Application of 2.0% **Phytofuse Renew**® demonstrated effective anti-pollution properties by preventing the accumulation of carbon on the skin.



**Figure 1.** Representative Images of Each Test Site Before and After Washing with Light Intensity Histograms of Each Site Before and After Washing. The green arrow indicates a shift back towards initial skin tones. The histogram threshold set from 0 (left) to 255 (right), where 0 reflects exclusively black pixels and 255 reflects exclusively white pixels.

## Carbon Removed from Skin Phytofuse Renew®



**Figure 2.** Carbon Removed from Skin After Washing. \* indicates significance ( $p \leq 0.05$ ) compared to Before Washing.

**Table 3.** One-way ANOVA Results of Histogram Analysis for Carbon Removed from Skin After Washing. \* indicates significance ( $p \leq 0.05$ ) between two conditions.

	Untreated Control vs Base Cleanser	Untreated Control vs 2.0% <b>Phytofuse Renew®</b>	Base Cleanser vs 2.0% <b>Phytofuse Renew®</b>
<b>P-value</b>	0.084	0.035*	0.042*

### Discussion

The results from this study demonstrate 2.0% **Phytofuse Renew®** provides carbon pollution protection as specified by micronized carbon residue. After carbon application, there was visibly less carbon present on the site treated with 2.0% **Phytofuse Renew®** compared to the Untreated Control and Base Cleanser test sites (Figure 1). Histogram analysis confirmed this lack of carbon accumulated on the skin via a smaller peak on the left which indicates the product was able to prevent the pollutant from depositing on the skin (Figure 1).

After washing, the test site treated with 2.0% **Phytofuse Renew®** visibly appeared cleaner and less carbon on the skin compared to the Untreated Control and Base Cleanser test sites (Figure 1). Histogram analysis confirmed 2.0% **Phytofuse Renew®** reduced carbon on the skin by 97%, whereas the Untreated Control and Base Cleanser exhibited reductions of 89% and 91%, respectively (Figure 2).

Taken together, these results indicate **Phytofuse Renew®** provides anti-pollution properties when added to personal care applications at recommended use levels. Collectively, **Phytofuse Renew®** reduces carbon accumulation on the skin and enhances carbon removal from the skin which improves the skin's protective barrier function and contributes to the appearance of healthier looking skin.

## References

1. Sharma AN, Patel BC. Laser Fitzpatrick Skin Type Recommendations. [Updated 2022 Mar 9]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan-. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK557626/>