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Tomorrow's Vision... *Today!*

# ABS Apple AHA's Efficacy Data

**Code:** 10286
**INCI Name:** Water & Propylene Glycol & Malic Acid & Pyrus Malus (Apple) Fruit Extract & Glycolic Acid & Lactic Acid & Citric Acid
**CAS #:** 7732-18-5 & 57-55-6 & 97-67-6 & 85251-63-4 & 79-14-1 & 50-21-5 & 77-92-9
**EINECS #:** 231-791-2 & 200-338-0 & 202-601-2 & 286-475-7 & 201-180-5 & 200-018-0 & 201-069-1

| Type of Study | Results |
|---|---|
| **Cellular Viability Assay** | ABS Apple AHA's exhibited positive results by increasing cell metabolism. The increase in fluorescent signal indicates an increase in cellular metabolism and viability post ABS Apple AHA's treatment. For these reasons, we can assume ABS Apple AHA's is suitable for cosmetic applications designed to increase cell viability and metabolism. |



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# Cellular Viability Assay Analysis

**Tradename:** ABS Apple AHAs

**Code:** 10286

**CAS #:** 7732-18-5 & 57-55-6 & 97-67-6 & 85251-63-4 & 79-14-1 & 50-21-5 & 77-92-9

**Test Request Form #:** 364

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

**Test Performed:**
Cellular Viability Assay

## Introduction

The cellular viability assay is useful for quantitatively measuring cell-mediated cytotoxicity, cell proliferation and mitochondrial metabolic activity. Increased metabolism in a cell indicates ample cellular respiration and adenosine triphosphate (ATP) production. ATP is the molecular energy of cells and is required in basic cell function and signal transduction. A decrease is ATP levels indicates cytotoxicity and decreased cell function while an increase in ATP levels indicates healthy cells.

The cellular viability assay was conducted to assess the ability of **ABS Apple AHAs** to increase cellular metabolic activity in cultured dermal fibroblasts.

## Assay Principle

The assay utilizes a nonfluorescent dye, resazurin, which is converted to a fluorescent dye, resorufin, in response to chemical reduction of growth medium from cell growth and by respiring mitochondria. Healthy cells that are in a proliferative state will be able to easily convert resazurin into resorufin without harming the cells. This method is a more sensitive assay than other commonly used mitochondrial reductase dyes such as MTT. An increase in the signal generated by resazurin-conversion is indicative of a proliferative cellular state.

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## Materials

| | | |
|---|---|---|
| **A.** | **Kit:** | PrestoBlue™ Cell Viability Reagent (Invitrogen, A13261) |
| **B.** | **Incubation Conditions:** | 37°C at 5% $CO_2$ and 95% relative humidity (RH) |
| **C.** | **Equipment:** | Forma humidified incubator; ESCO biosafety laminar flow hood; Light microscope; Pipettes |
| **D.** | **Cell Line:** | Normal Human Dermal Fibroblasts (NHDF) (Lonza; CC-2511) |
| **E.** | **Media/Buffers:** | Dulbecco's Modified Eagle Medium (DMEM); Penicillin-Streptomycin (50U-50mg/mL); Fetal Bovine Serum (FBS); Phosphate Buffered Saline (PBS) |
| **F.** | **Culture Plate:** | Falcon flat bottom 96-well tissue culture treated plates |
| **G.** | **Reagents:** | PrestoBlue™ reagent (10X) |
| **H.** | **Other:** | Sterile disposable pipette tips |

## Methods

Human dermal fibroblasts were seeded into 96-well tissue culture plates and allowed to grow to confluency in complete DMEM. A 10-fold serial dilution was performed resulting in **ABS Apple AHAs** concentrations on 1%, 0.1%, and 0.01% in complete DMEM and incubated with fibroblasts for 24 hours.

Ten microliters of viability reagent was added to 90µL of cell culture media in culture wells.

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## Results

The data obtained from this study met criteria for a valid assay and the controls performed as anticipated.

**ABS Apple AHAs** at all concentrations is able to increase cellular metabolism compared to the control.

Cellular metabolism results are expressed as a percentage of the control.

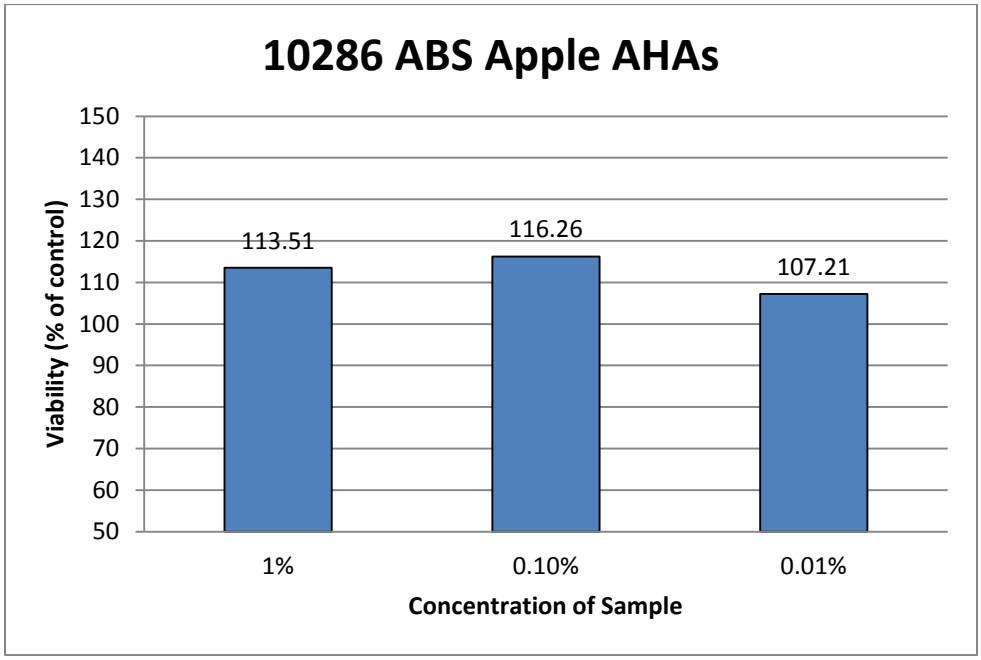


**Figure 1:** Cellular Metabolism of **ABS Apple AHAs**-treated fibroblasts expressed in terms of percent of control.

## Discussion

As shown in figure 1, **ABS Apple AHAs** exhibited positive results by increasing cell metabolism. The increase in fluorescent signal indicates an increase in cellular metabolism and viability post **ABS Apple AHAs** treatment. For these reasons, we can assume **ABS Apple AHAs** is suitable for cosmetic applications designed to increase cell viability and metabolism.