

# Establishment and application of a novel experimental model of stratum corneum carbonylation induced by UV and sebum

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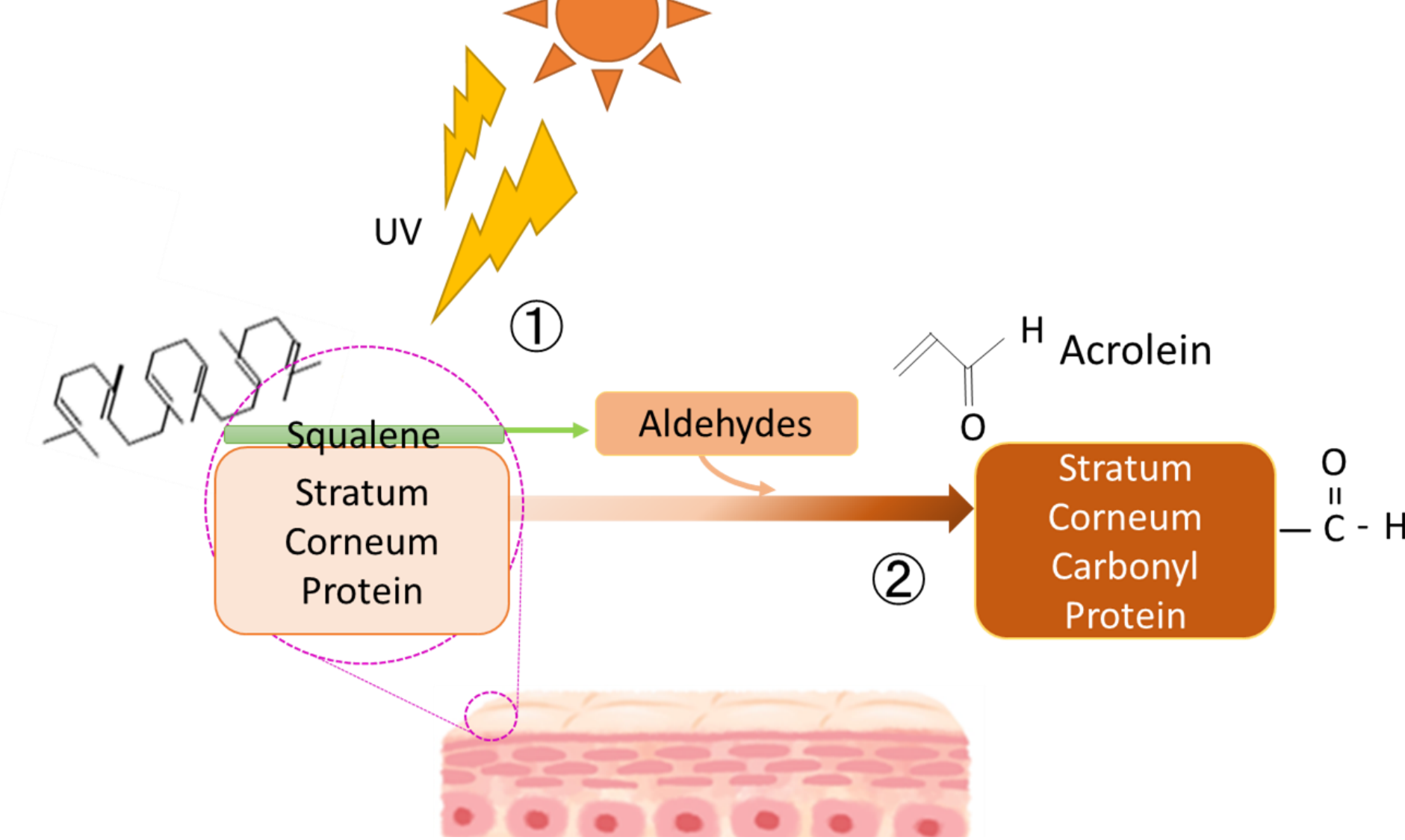
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## Conclusion

We established an experimental stratum corneum carbonyl protein (SCCP) induction model mimicking oxidative modification at the skin surface using a tape-stripped stratum corneum (SC) irradiated with ultraviolet (UV) + squalene (SQ). This experimental system made it possible to evaluate the efficacy of cosmetic ingredients in suppressing SCCP to shield healthy skin.

## Introduction

Carbonyl protein (CP) is formed by various oxidative stimuli, including UV radiation.

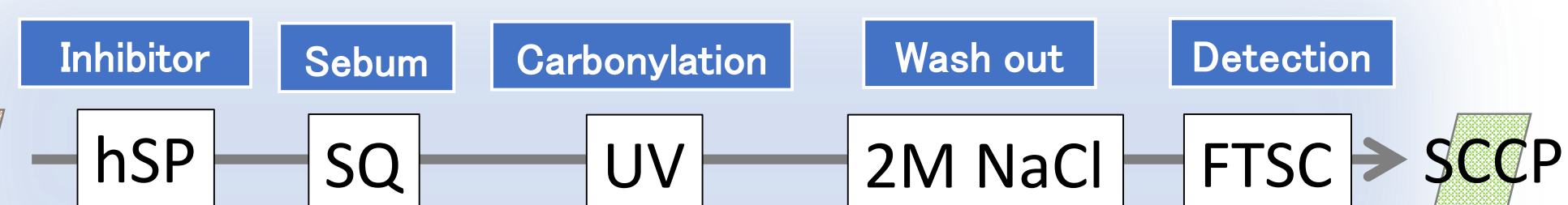


- ① Unsaturated lipids (such as SQ) → Lipid peroxides → Degradation → Various aldehydes (such as acrolein (Acr))
- ② Aldehydes → Attack → SC proteins → SCCP

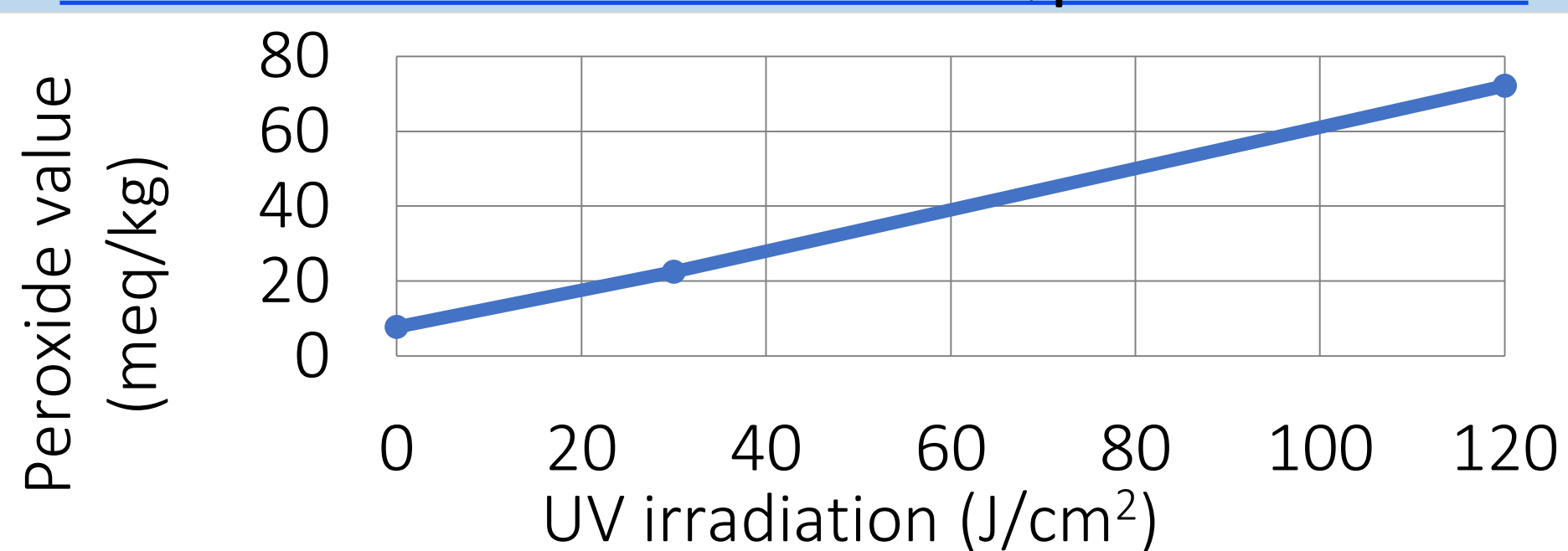
We need an experimental system ① + ②

## Materials and Methods

- Materials
  - Hydrolyzed soy protein (hSP)
  - : Mixture of peptides (Average molecular weight 700 Da, Many basic amino acid residues such as Lys, Concentration 20%)
  - TOFUPRO® U, IKEDA CORPORATION
- SC collection
  - The inside of the upper arms by tape-stripping
- Carbonylation (induce SCCP)
  - UV (LAX-C100 solar simulator (Asahi Spectra Co., Ltd.), SQ + UV, Acr
- Evaluation of SCCP
  - Fluorescein-5-thiosemicarbazide (FTSC), EVOS-FL fluorescence microscope (Thermo Fisher Scientific)
- Wash out
  - Soaking the SC in 2 mol/L NaCl

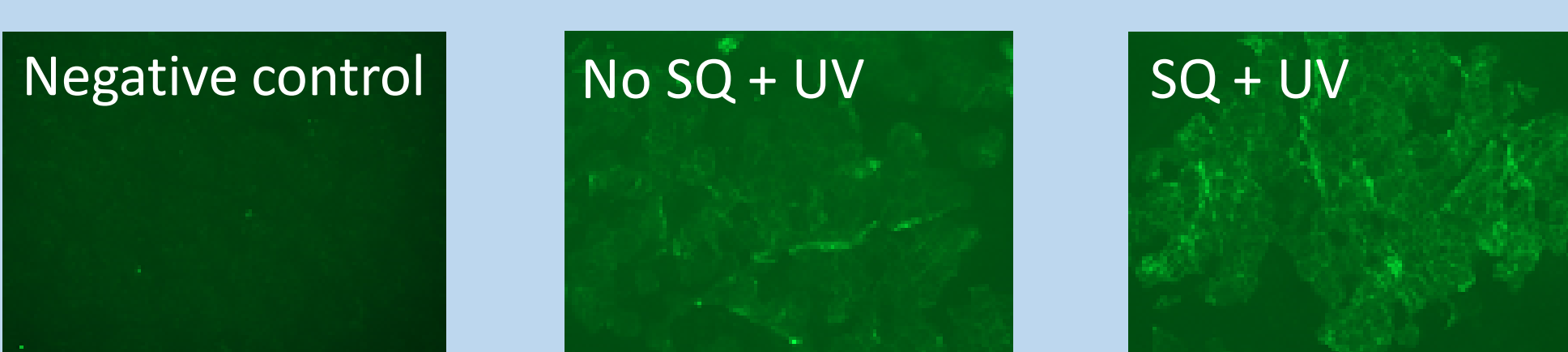


### Effect of UV irradiation on SQ peroxide value



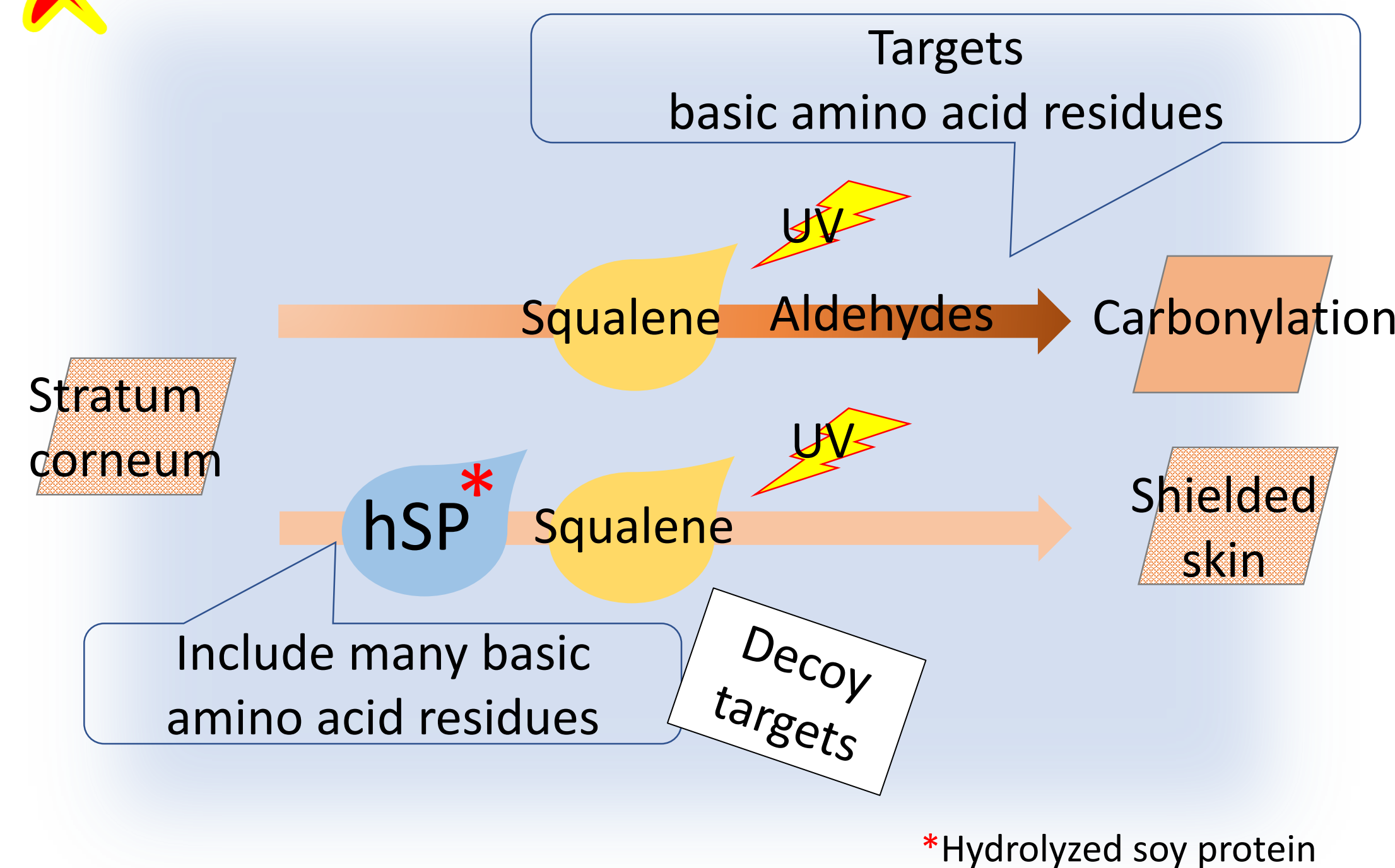
SQ was irradiated with different amounts of UV. Peroxides were consistently produced in SQ irradiated with UV.

### Detection of carbonyl protein by Acr antibody



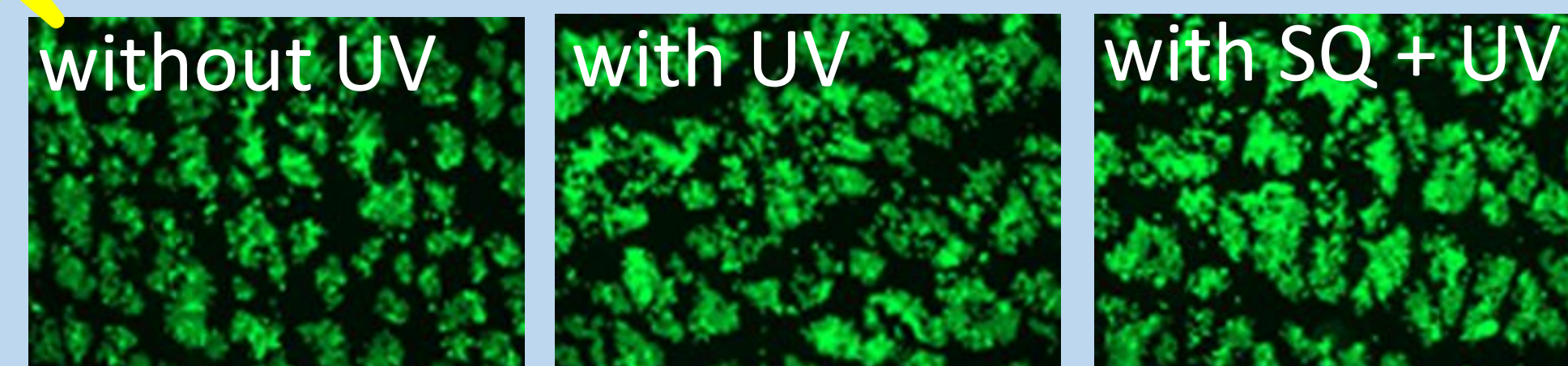
SC was treated without or with SQ, then irradiated with UV. Acr adduct was generated in the SC treated with SQ + UV.

We established an experimental

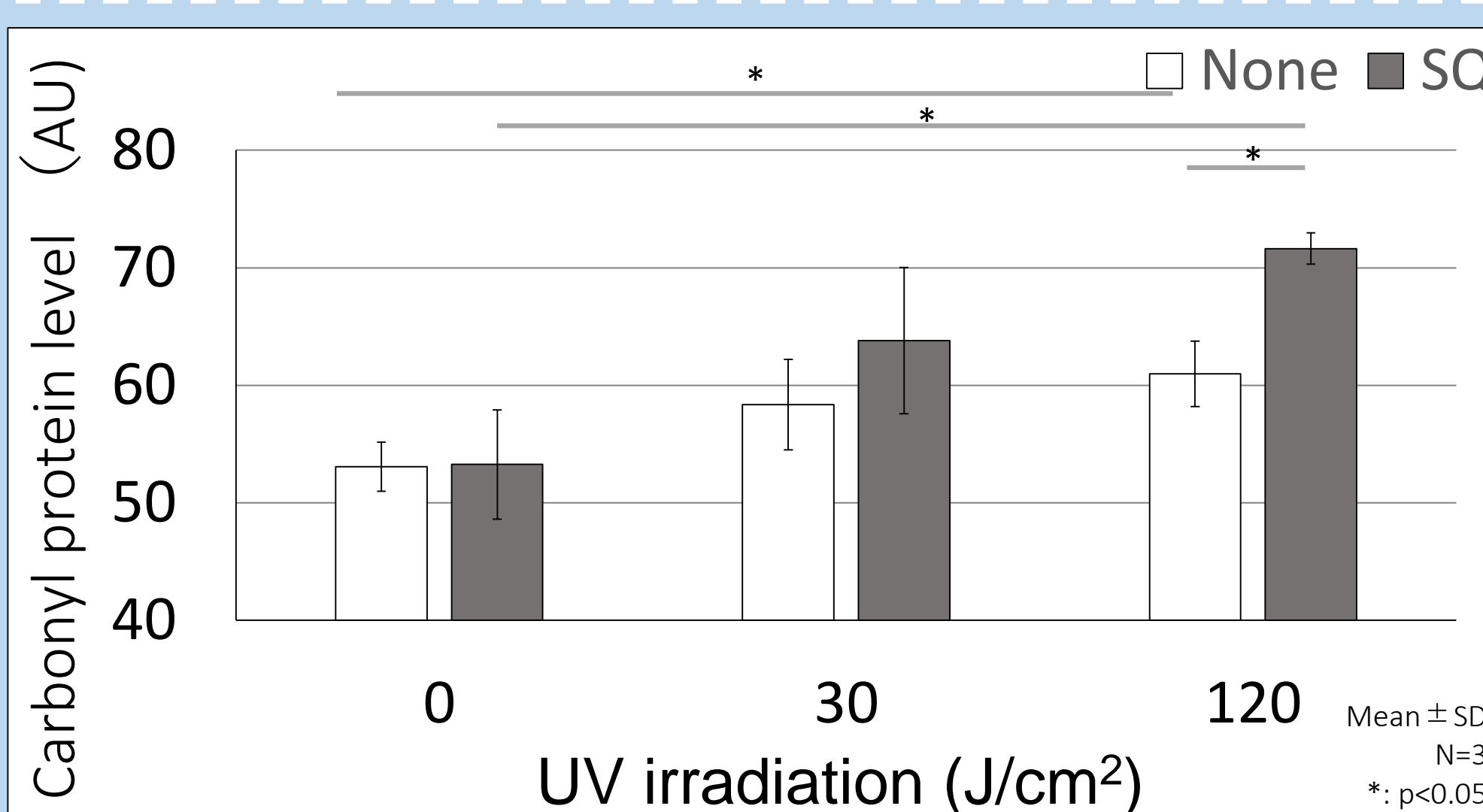


## Results

### Promotion of UV-induced SCCP by SQ

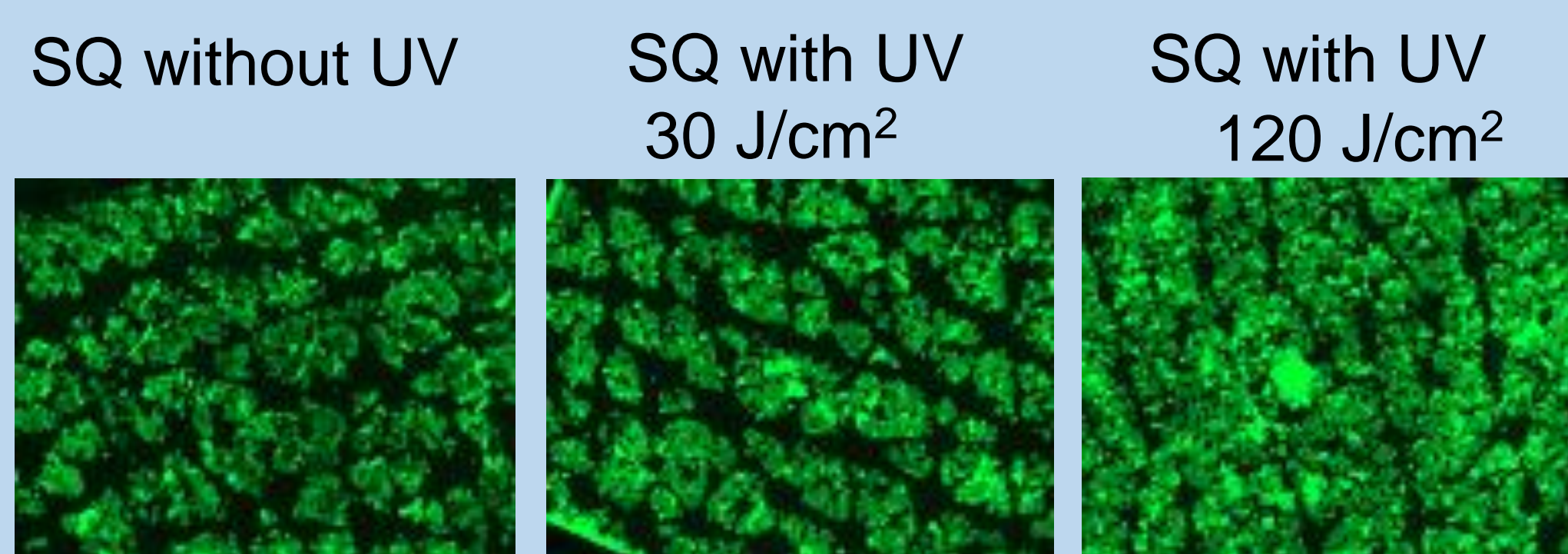


Typical images of SCCP following treatment without UV, with 120 J/cm<sup>2</sup> UV, or 120 J/cm<sup>2</sup> UV + SQ. UV promoted greater SCCP induction in the presence of SQ.



SCCP was successfully induced by UV irradiation in a dose-dependent manner. We established a novel experimental model of SC carbonylation induced by UV + sebum component.

### Induction of SCCP by UV-irradiated SQ



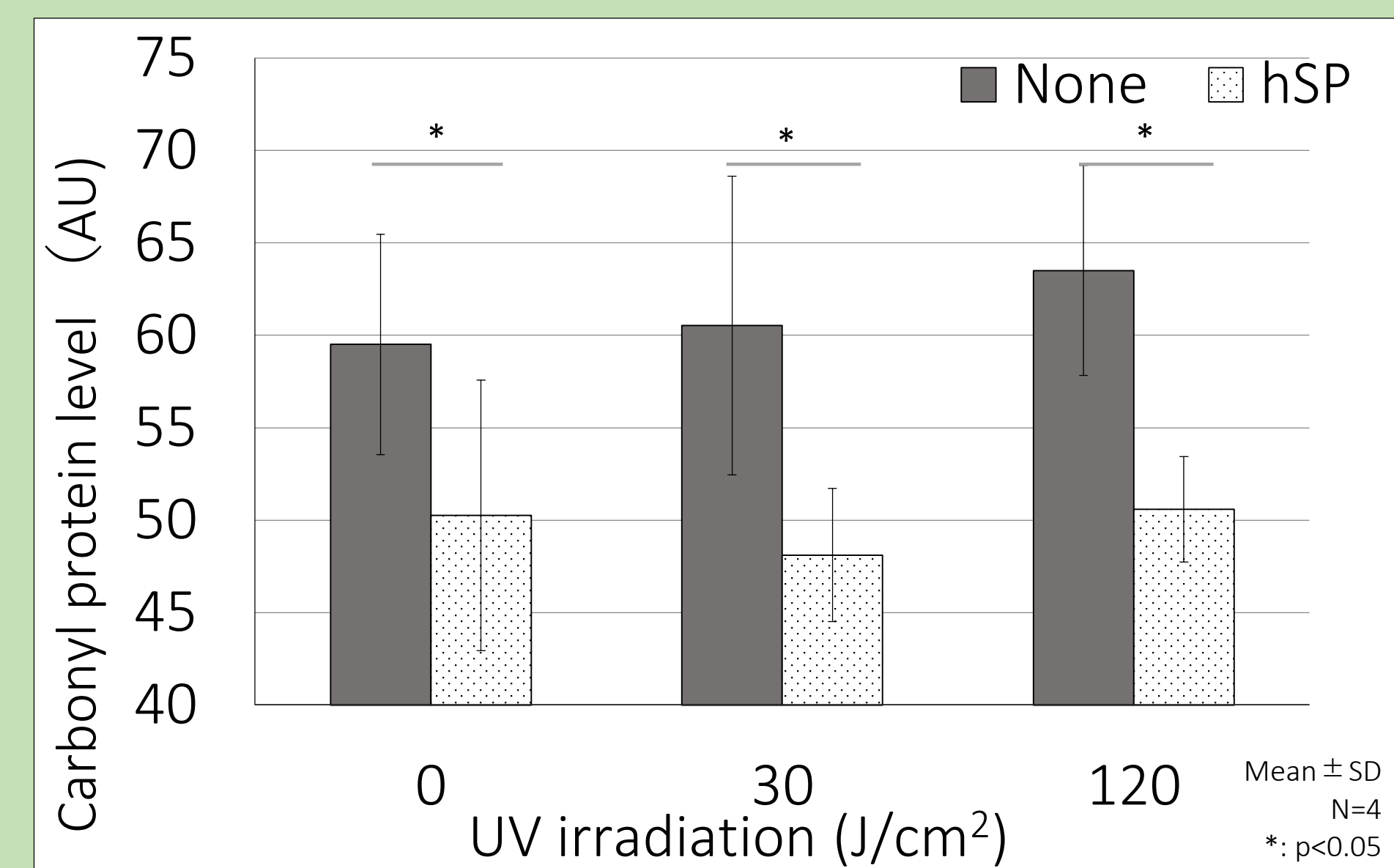
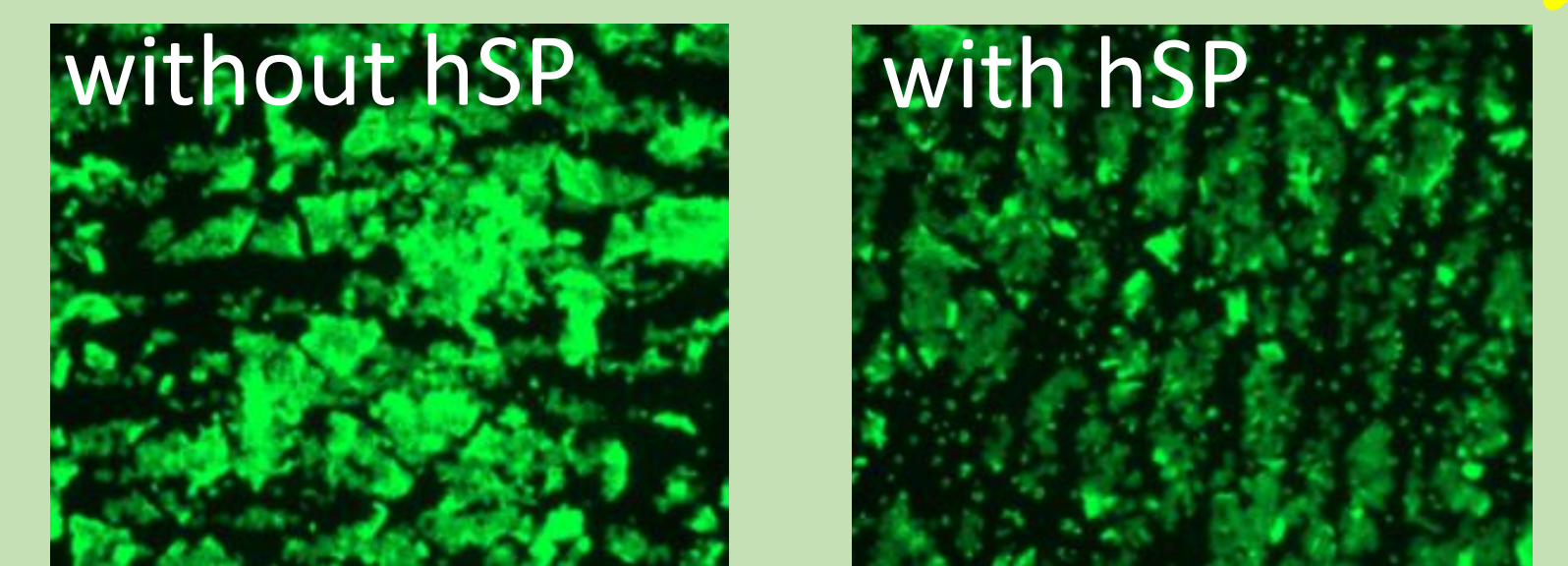
The SCCP level was increased in a dose-dependent manner by application of UV-irradiated SQ.

### Inhibitory effect of hSP on Acr-induced SCCP induction



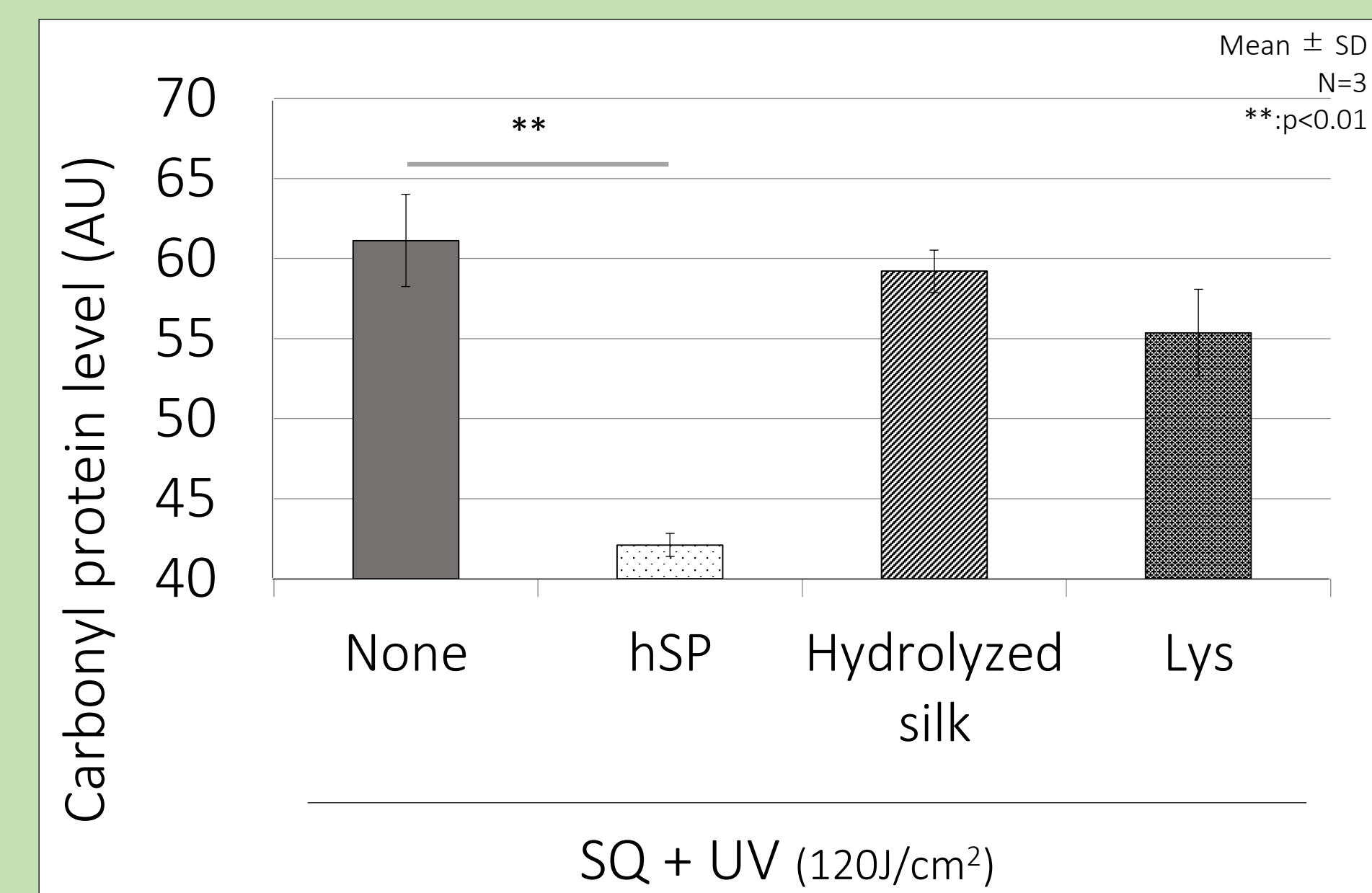
SC was treated with deionized water or hSP, then treated without or with 1 mmol/L Acr. hSP showed an excellent inhibitory effect against SCCP induction by Acr.

### Inhibitory effect of hSP on UV-induced SCCP induction in the presence of SQ



SC was treated without or with hSP, then irradiated with UV + SQ. hSP application to the tape-stripped SC prior to UV + SQ resulted in significant inhibition of SCCP generation.

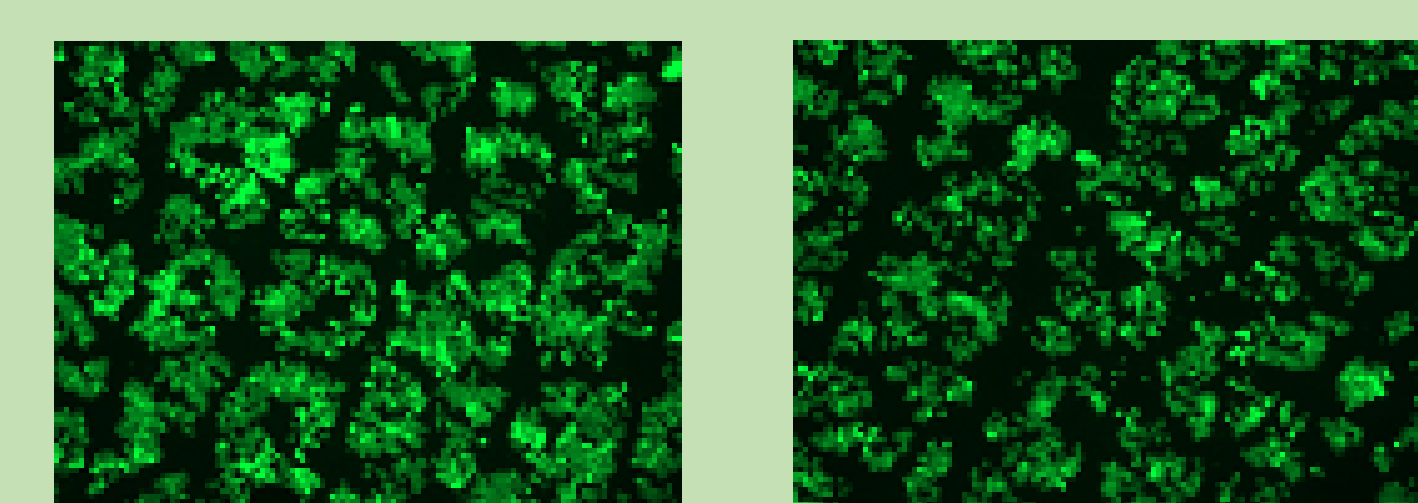
### Inhibitory effect of cosmetic ingredients on UV-induced SCCP induction in the presence of SQ



hSP exhibited an excellent inhibitory effect against the SCCP induction as compared with hydrolyzed silk or L-Lys.

### Loss of the inhibitory effect of hSP on SCCP induction by SQ + UV following removal of hSP prior to SCCP induction

	Pre-wash	Post-wash
hSP	+	+
Wash	+	-
SQ + UV (30J/cm²)	+	+
Wash	-	+



Removal of adsorbed hSP prior to SCCP induction (pre-wash) eliminated the inhibitory effect of hSP against UV-induced SCCP induction in the presence of SQ.