

Grating:1800 gr/mm

Confocal pinhole:Open, 150 μm

Spectral range:400–1800 cm^{-1}

Spectrometer center:1100 cm^{-1}

Integration time:10 s/point

Accumulation:2 times

Scanning mode:Line mapping (SWIFT)

Step size:20 μm

Autofocus:On, AF period = 0.5 min

4.4.3 Skin Model Preparation

Ex vivo porcine skin was purchased from Pizhou Dongfang Breeding Co., Ltd., stored at -25°C .

Upon use, the skin was thawed at room temperature, cut into $1.5\text{ cm} \times 1.5\text{ cm}$ pieces (thickness~1 mm), rinsed twice with normal saline, and randomly divided into 4 groups (3replicates per group). The skin pieces were placed on moistened cotton pads in 60 mm petri dishes (40 μL saline on a $3 \times 3\text{ cm}$ cotton pad) to keep the skin surface (stratum corneum) facing upward.

4.4.4 Experimental Groups

The following four groups were established, each with 3 replicates.

Blank group: Untreated ex vivo skin. This group provides the skin's own Raman spectral baseline, serving to exclude interference from endogenous skin components with the characteristic peaks of the target ingredients. By comparing the spectra of the blank group and the treated groups, the characteristic peaks of exogenous components can be clearly identified, and it can also serve as a reference for normalization (e.g.,Amide I peak).

Control group:Ex vivo skin treated with essence only (no beauty device). This group simulates the natural absorption process of the essence on the skin surface.

Positive control group:10% DMSO solution. This group is used to verify that the experimental system (skin model, sectioning, Raman detection) can detect the absorption of a known penetration enhancer, thereby ensuring the sensitivity and

reliability of the experimental method.

Test group: Ex vivo skin treated with essence followed by irradiation with the beauty device. This group is used to evaluate the absorption-promoting effect of the beauty device on the essence.

4.4.5 Pre-treatment for Different Groups

Blank group: Skin left at room temperature for 4 min, then incubated in a 37°C water bath for 1 h, and immediately frozen at -25°C.

Control group: 20 µL essence evenly applied to skin surface, left at room temperature for 4 min, then incubated at 37°C for 1 h, and frozen.

Test group: 20 µL essence applied, then the beauty device (function 2, level 5) was applied vertically with static contact, lightly pressed to cover the skin completely for 4 min, then incubated at 37°C for 1 h, and frozen.

Positive control group: 20 µL 10% DMSO applied, left at room temperature for 4 min, incubated at 37°C for 1 h, and frozen.

4.4.6 Cryosectioning

Frozen samples were equilibrated in the cryostat chamber for 15 min. They were embedded in OCT with the stratum corneum facing outward. After freezing, sections were cut perpendicular to the skin surface (vertical sections). Trimming was performed until the skin tissue was exposed. Section thickness: 25 µm. The first 3 sections were discarded, and the next 3 consecutive sections were collected and placed on ice.

4.4.7 Raman Spectral Acquisition

The device was preheated for 15 min and calibrated with a silicon wafer (520.7cm^{-1}).

For the essence and DMSO samples: The sample was placed on the stage, visualized with a 50× objective, and a single-point SWIFT mode spectrum was acquired.

For skin sections: After thawing at room temperature for 5 min, the section was placed on the stage. Using a 10× objective, the skin cross-section was located. The objective was switched to 50× and focused. The laser spot was placed on the skin tissue (avoiding OCT). Points were measured at depths of 0, 20, 40, and 60 µm from the skin surface (stratum corneum edge) with a step size of 20 µm.

4.4.8 Data Processing and Statistical Analysis

4.4.8.1 Data Preprocessing

(equipped with negative ions, microcurrent, and red/yellow light) on an essence. Four groups were established: blank (untreated ex vivo skin), control (essence only), test (essence + beauty device treatment for 4 min), and positive control (10% DMSO). Raman spectra were acquired at depths of 0-60 μm .

The positive control (10% DMSO) was used to verify the experimental system. The C-S peak of DMSO was detected up to 40 μm depth. According to literature, pure DMSO molecules exhibit a doublet at ~ 670 and $\sim 698\text{ cm}^{-1}$ due to different aggregation states (dimers, trimers). However, when DMSO is mixed with water, solvent effects homogenize the C-S bond vibration, causing the original doublet to merge into a single broad peak - consistent with our observation.

The test group tracked the characteristic peak of the active ingredient ($\sim 1600\text{ cm}^{-1}$) at various depths. The results showed that the beauty device extended the effective absorption depth from approximately 0 μm (control) to 40 μm . The absorption-promoting effect likely arises from the synergistic action of multiple functions: negative ions (charge repulsion to drive negatively charged active ingredients deeper), microcurrent (increased cell membrane permeability), and red/yellow light (photothermal effect to accelerate microcirculation and uniform distribution). This is consistent with reported physical penetration enhancement techniques such as iontophoresis and sonophoresis for small hydrophilic molecules.

7 Conclusion

This Raman spectroscopic depth-scanning study demonstrated that without promotion, the active ingredient of the essence remains mainly on the skin surface (0 μm). With the home beauty device, the absorption depth reached 40 μm . Thus, the home beauty device promotes the absorption of the essence without causing discernible skin damage.

8 References

- [1] OUYANG Shunli;ZHANG Mingzhe;HU Qing-cheng, et al .Hydrogen Bonding Effect on the Surface Tension and Viscosity of DMSO Aqueous Solutions Studied by Raman Spectroscopy[J].*Spectroscopy and Spectral Analysis*, 2018, 38(9): 2778.