

In Vivo Raman Study of Retinol Skin Penetration for Anti-Aging Applications

■ Abstract

This application note demonstrates how the IN VIVO RAMAN ANALYZER “RZ660” can be used to evaluate the skin penetration of Retinol, a key anti-aging ingredient. The study investigates the depth of Retinol penetration in human skin over time, with results presented visually to provide solid scientific & marketing support for cosmetic efficacy claims.

■ Background and challenges

As consumers increasingly demand cosmetics backed by scientific validation, understanding how active ingredients penetrate the skin has become a key factor in evaluating product efficacy. Traditional analytical methods, such as tape stripping and Franz diffusion cells, are widely used but have notable limitations. Tape stripping is a destructive technique restricted to the stratum corneum, while Franz diffusion testing is performed in vitro and cannot fully reproduce real skin conditions. These approaches also lack spatial and molecular resolution, limiting their ability to

describe penetration mechanisms in detail. In contrast, *in vivo* Raman spectroscopy provides a non-destructive and label-free solution for studying the molecular behavior of active ingredients directly in human skin. This technique enables visualization of the skin's structure, monitoring of active diffusion through different layers, and correlation between penetration profiles and cosmetic performance, offering a more comprehensive and realistic understanding of skin interactions.

■ Methodology

| Section | Description |
|-------------------------|--|
| Sample & Application | Retinol essence lotion applied at a dosage of 2 mg/cm ² the inner forearm region. |
| Measurement Time Points | Skin measurements were taken on control (Blank skin) areas and at 1 h, 2 h, and 4 h after product application. |
| Depth Range | Measurements were acquired over a depth range from -20 µm to 100 µm, taking the skin surface as the 0 µm reference point. |
| Instrument | IN VIVO RAMAN ANALYZER “RZ660” used for testing (see Figure 1). |
| Instrument Features | Equipped with a remote probe allowing easy access to various body areas. Single acquisition covering 600–3800 cm ⁻¹ provides data on ingredient penetration and skin structure changes. |



Figure 1: IN VIVO RAMAN ANALYZER "RZ660"

Results

Figure 2 presents the characteristic Raman spectra of human skin (blue) and Retinol (red). The skin spectrum displays distinct bands corresponding to key components such as keratin, lipids, and water. Retinol shows notable spectral differences compared to human skin, and these characteristic signals allow for non-destructive monitoring of Retinol penetration in the skin.

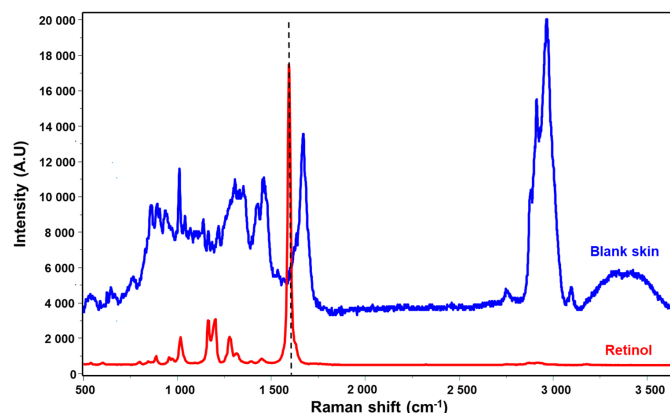


Figure 2: Raman Spectra of Blank Human Skin and Retinol.

Figure 3 shows Raman depth imaging of human skin before (Blank skin) and at 1, 2, and 4 hours after application of Retinol essence. The images reveal the distribution and progressive penetration of Retinol within the stratum corneum and deeper skin layers. Penetration is visualized at the micron scale, with warmer colors indicating higher Retinol concentration. In addition to tracking Retinol diffusion, these measurements simultaneously provide information on skin hydration and barrier properties.

Figure 4.a shows 2D and 3D Raman imaging of Retinol penetration after 4 hours of application; the black line indicates the stratum corneum, showing that Retinol has reached the active epidermis. Figure 4.b shows Skin hydration mapping, providing quantitative evidence of cosmetic effects such as moisturizing, anti-aging, and brightening.

Conclusion

The IN VIVO RAMAN ANALYZER “RZ660” enables precise, non-invasive assessment of cosmetic ingredient behavior in human skin. Using 2D and 3D Raman imaging, it is possible to track the penetration of actives like Retinol through the stratum corneum over time, while simultaneously evaluating skin hydration and barrier function. This provides solid scientific evidence to support efficacy claims such as moisturizing, anti-aging, and brightening. Beyond validation, the generated images serve as powerful marketing tools, visually demonstrating product performance and enhancing brand credibility. Overall, this approach bridges research, formulation optimization, claim substantiation, and marketing communication, making it a valuable tool in cosmetic development.

info.sci@horiba.com

USA: +1 732 494 8660
France: +33 (0)1 69 74 72 00
Germany: +49 (0) 6172 1396 0
UK: +44 (0)1604 542 500

Italy: +39 06 51 59 22 1
Japan: +81(75)313-8121
China: +86 (0)21 6289 6060
India: +91 (80) 4127 3637

Singapore: +65 (6) 745-8300
Taiwan: +886 3 5600606
Brazil: +55 (0)11 2923 5400
Other: +33 (0)1 69 74 72 00

Raman imaging provides precise analysis of how cosmetic ingredients interact with the skin, showing active ingredient penetration, such as Retinol, through the epidermis. Skin hydration mapping quantifies product effects, supporting claims like “deeply moisturizes,” “reduces wrinkles,” or “improves skin radiance.” Beyond scientific validation, 2D and 3D images serve as compelling marketing tools, visually demonstrating efficacy and strengthening brand credibility.

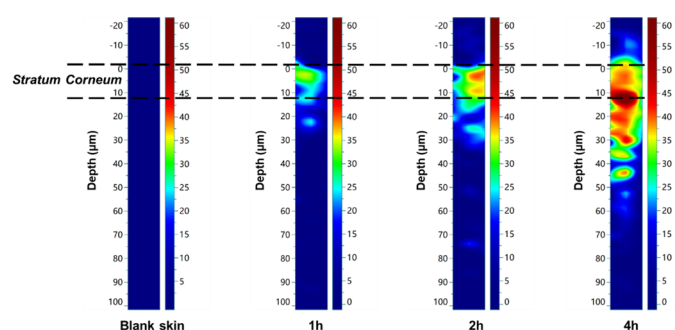


Figure 3: Raman images showing Retinol penetration into human skin at 1, 2, and 4 hours after application, compared with untreated (blank) skin.

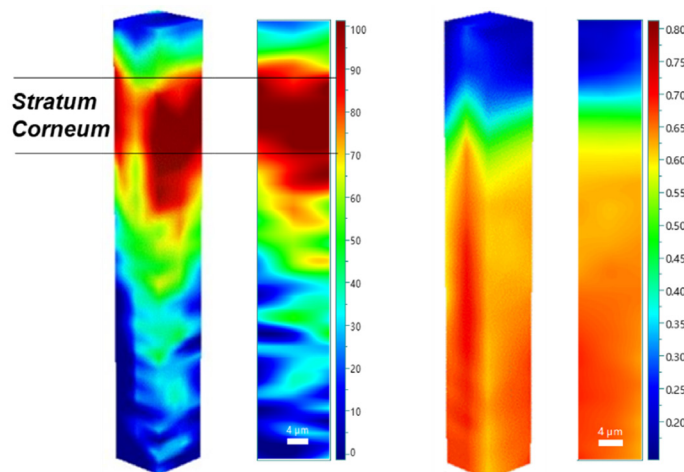


Figure 4 : 3D and 2D Raman images showing (a) Retinol distribution after 4 hours of application and (b) skin water content.

www.horiba.com/scientific