

CHARACTERIZATION OF A SYNERGISTIC MICROGEL COMPLEX THAT IMPROVES ACNE TREATMENT EFFICACY

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ABSTRACT

A novel synergistic microgel complex has been developed to provide a holistic acne treatment—delivering anti-acne efficacy without the unwanted effects such as irritation, flaking, compromising barrier function or inducing oxidative stress. While the effects of oxidative stress are not immediately apparent to the patient, its cumulative effects are detrimental to overall skin health. The microgel complex is a multiple-phase system containing complementary ingredients that provide sebum-dissolving properties and enhanced delivery of anti-acne agents. Several methods were used to characterize the technology:

Mechanisms of Action and Delivery

- The microgel complex was characterized by light microscopy, showing that the median diameter of a microgel is ~10 µm.
- The microgel complex with 2% salicylic acid was analyzed by HPLC. Results show that the microgel contains a higher concentration of salicylic acid compared to the external phase and the microgel consists of the sebum solubilizing ingredients described below. These properties may contribute to the enhanced delivery of salicylic acid into the skin for keratolytic effects.
- Sebum solubilization: *In vitro* investigations with model sebum compositions showed that components of the microgel complex solubilize sebum. This sebum solubilization property has potential to help with treating microcomedones and open comedones by improving sebum flowability and skin penetration.
- Skin penetration in human subjects was evaluated to assess the distribution of salicylic acid as a function of time. The formulation containing the microgel complex and 2% salicylic acid demonstrated enhanced delivery versus a formulation with 2% salicylic acid alone.
- In vitro* antimicrobial activity against *Propionibacterium acnes* was evaluated, showing that 2% salicylic acid microgel complex kills 99.9% of *P. acnes* in 15 seconds.

Skin Protective Effects

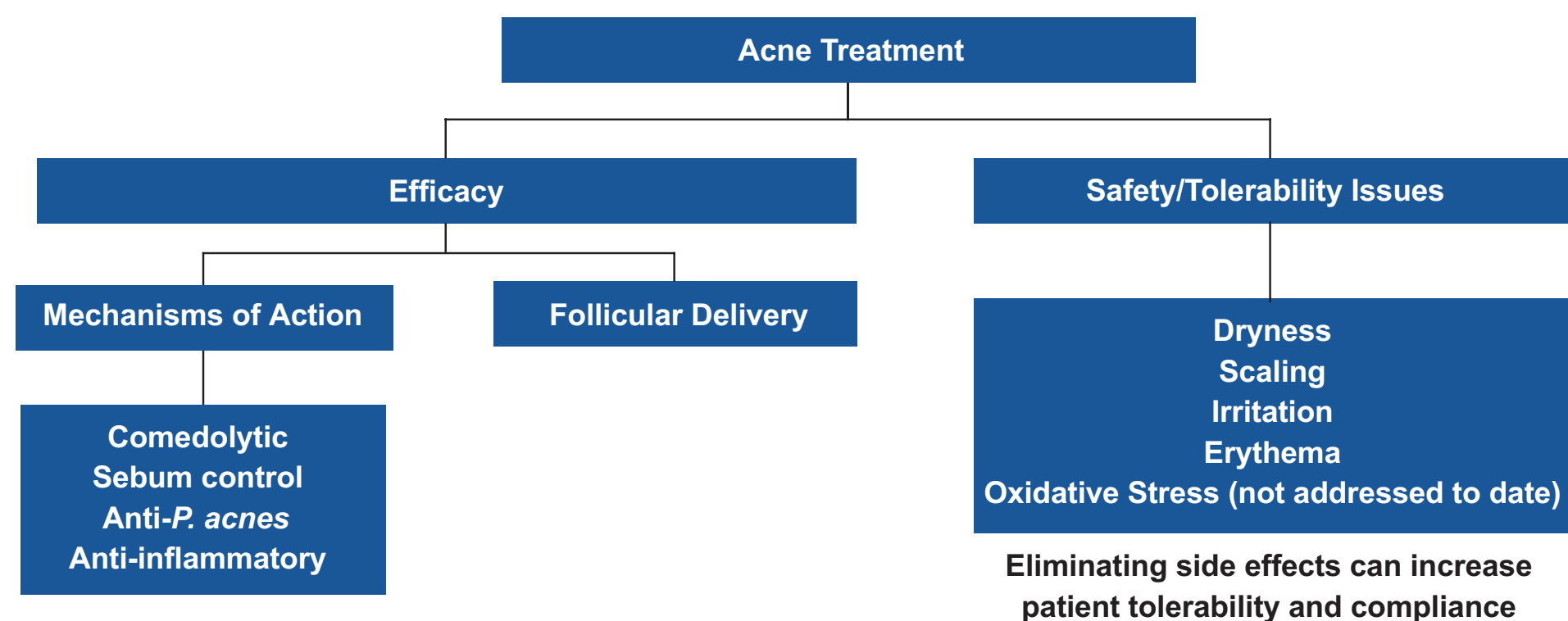
- Skin barrier effects: In a clinical study (n=13), skin conductance and TEWL results demonstrated that the formulation with the microgel complex significantly improves skin moisture content and does not compromise the barrier function of the skin.
- Oxidative Stress: Reactive oxygen species (ROS) were measured on human subjects by *in vivo* chemiluminescence and *ex vivo* H₂O₂ production. Results demonstrate the 2% salicylic acid microgel complex does not induce oxidative stress on the skin, while 10% BPO induces significant amounts of oxidative stress. While the effects of oxidative stress are not immediately apparent to the patient, its cumulative effects can result in accelerated aging of the skin.

The characterization of the novel microgel complex demonstrates the technology has unique sebum solubilization properties, enhanced skin penetration, and fast-acting anti-*P. acnes* activity, without causing oxidative stress or compromising the skin barrier function. Clinical studies with this microgel complex confirm its safety and efficacy in treating acne.

INTRODUCTION

Acne is a follicular disease caused by excessive sebum production, clogging of the follicular duct and when present, proliferation of pathogenic bacteria *P. acnes*. Major efforts have been dedicated to improving active delivery to target sites (eg, pilosebaceous unit) and reducing irritation and dryness caused by acne treatments. Side effects that patients commonly experience with acne treatments such as irritation, erythema, and scaling often lead to lower patient compliance. In addition, these treatments often take several days to weeks before the patient or clinician can note a clinical improvement. There is a need for efficacious acne treatments that can deliver actives to the target site without causing undesirable effects, ultimately improving patient compliance and clinical outcomes.

Figure 1 - Aspects of Acne Treatments



In this poster, we describe the unique characteristics of a synergistic microgel complex that functions through multiple mechanisms of action, improves delivery, and addresses safety/tolerability issues often found in acne treatments (Figure 1). Clinical studies demonstrate that this microgel complex provides rapid target lesion treatment and acne prevention without irritation or dryness.

METHODS

Microscopy - Approximately 50 mg of product was placed on a glass slide to determine the dimensions of the microgels using a light microscope (Nikon Polarizing Light Microscope).

Composition of the Microgel Complex - A composition containing 2% salicylic acid and microgel complex was analyzed using reverse-phase isocratic high performance liquid chromatography (HPLC).

Sebum Solubilization - Model sebum compositions were formulated with triglycerides, wax esters, squalene, free fatty acids, cholesterol esters and cholesterol based on the percentages found in human sebum. The investigative ingredients were then added to the model sebum and measured on an opacity scale at baseline, 2 hours, and 6 hours.

Skin Penetration in Human Subjects - Fluorescence spectroscopy readings¹ were taken on the volar forearm at baseline and 20 minutes and 60 minutes post-product application to evaluate the time dependent penetration profile. Fluorescence spectroscopy evaluations were also done on each test site after 5, 10 and 12 D-square tapes.

In vitro Antimicrobial Efficacy Against Propionibacterium acnes - The Bacterial Contact Time Kill (BCT) test was used to determine the product contact time required to show cidal activity against *P. acnes*.

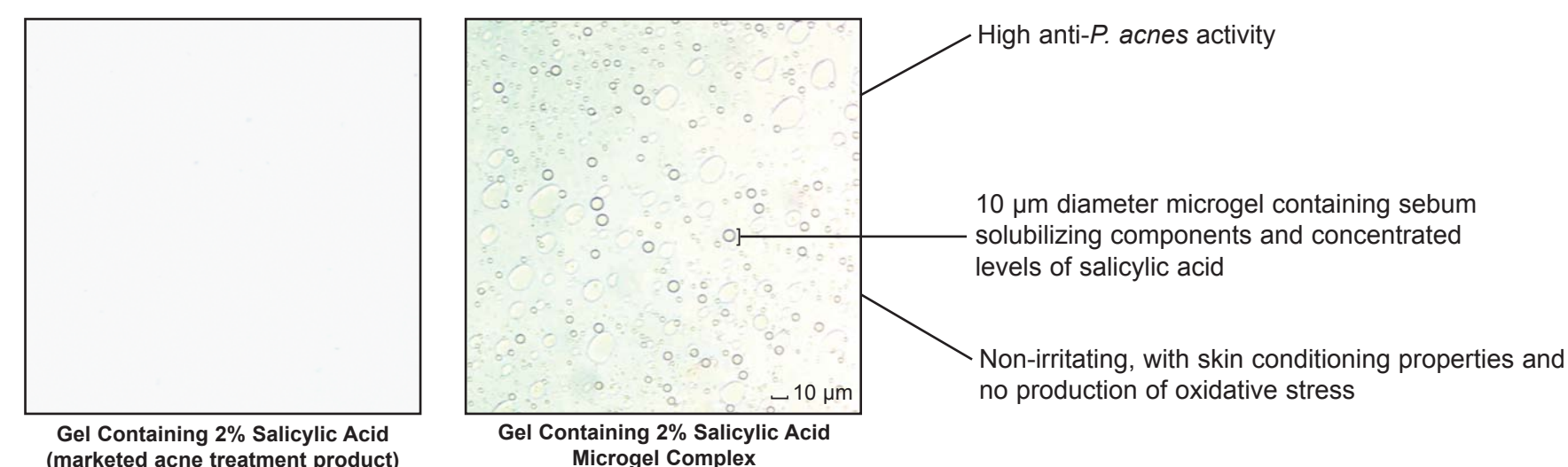
Skin Barrier Properties in Human Subjects - Skin Conductance (Skicon®) and Transepidermal Water Loss (TEWL) were used to evaluate skin moisture and barrier properties on the volar forearms of 13 volunteers.

Oxidative Stress - The amount of reactive oxygen species (ROS) produced on the skin by test products was determined *in vivo* using a chemiluminescence method² and *ex vivo* using a fluorescent cellular probe, which when taken up by the cells fluoresces upon oxidation by cellular-generated peroxy radicals.

RESULTS

MICROSCOPE IMAGE OF MICROGEL COMPLEX

Figure 2 - Comparison of Microscope Images of a Gel Containing the Novel 2% Salicylic Acid Microgel Complex versus a 2% Salicylic Acid Marketed Acne Treatment Gel (189x magnification).



As shown through the microscope images in Figure 2, the marketed 2% salicylic acid acne treatment gel is a single phase system, while the gel containing the 2% salicylic acid microgel complex is a multiple-phase system. The 10 µm diameter microgels contain sebum solubilizing components and concentrated levels of salicylic acid.

COMPOSITION OF THE MICROGEL COMPLEX

HPLC analysis of salicylic acid in the microgel complex composition demonstrated that the microgel contains 1.6-fold higher salicylic acid versus the external phase. The microgel also consists of the sebum solubilizing ingredients. These properties may contribute to the enhanced delivery of salicylic acid into the skin for keratolytic effects.

SEBUM SOLUBILIZATION

Figure 3 - Effect of Microgel Components on Sebum Dissolution Rate

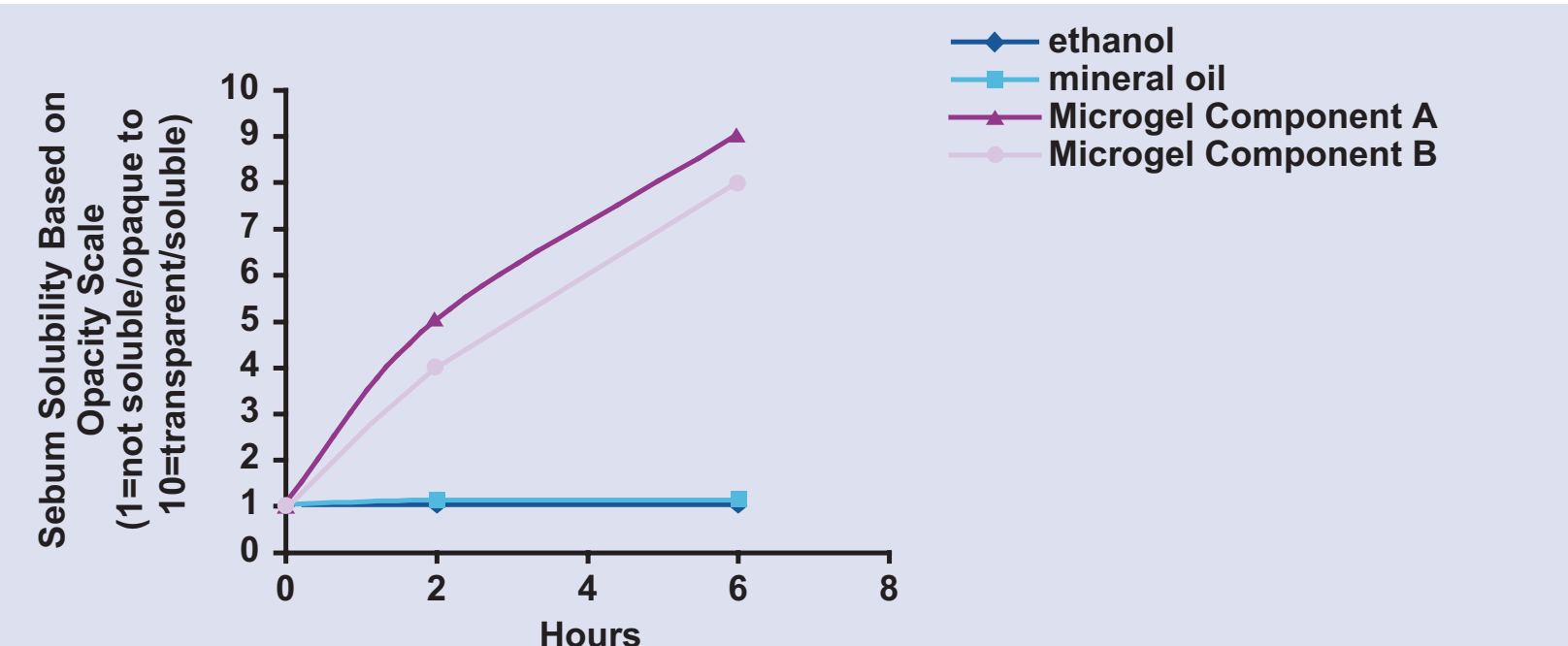
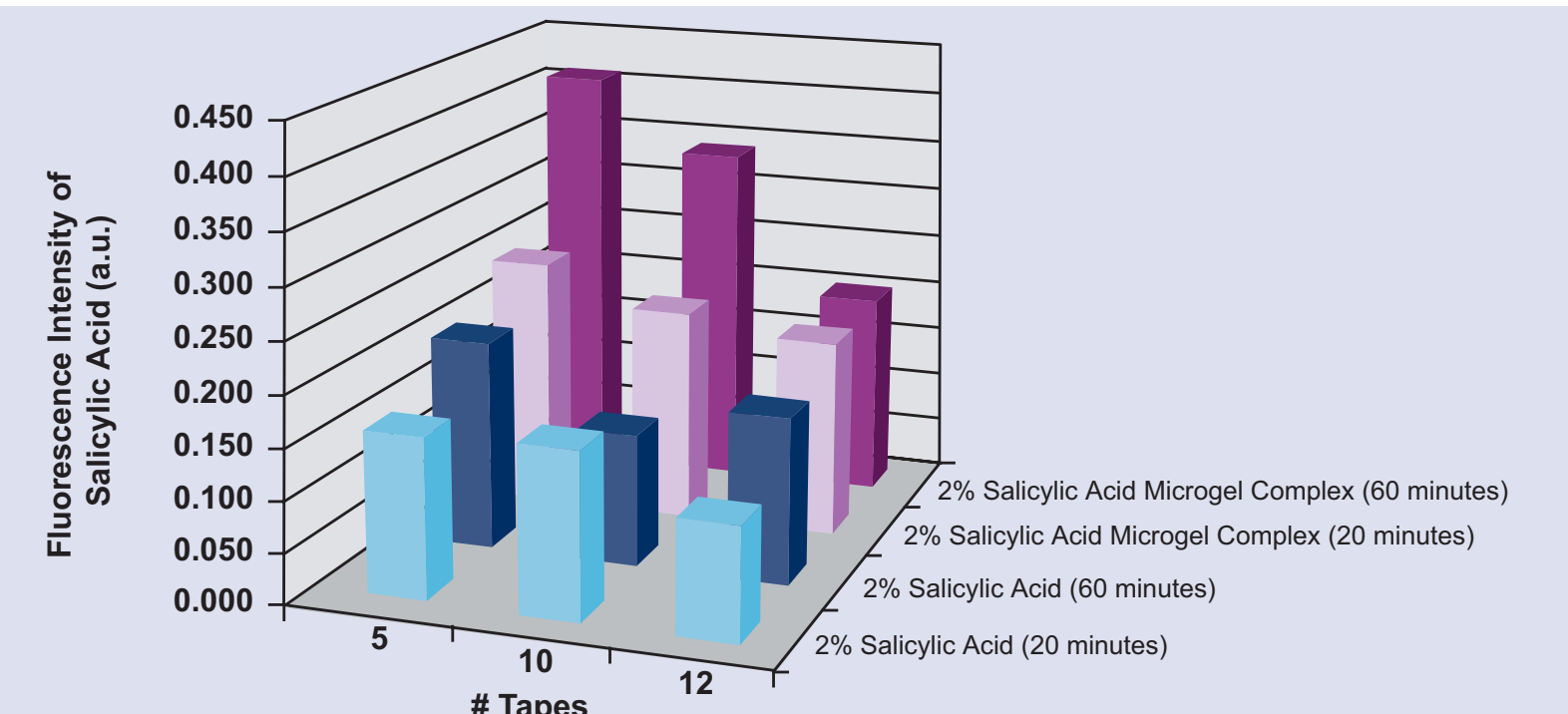


Figure 3 shows that the components of the microgel complex solubilize sebum over time. In contrast, ethanol and mineral oil do not solubilize the model sebum, as indicated by the opacity at all time points. This sebum solubilization property has potential to help with treating microcomedones and open comedones by improving sebum flowability and skin penetration.

SKIN PENETRATION OF SALICYLIC ACID IN VIVO

Figure 4 - Effects of the Novel Microgel Complex on Salicylic Acid Distribution as a Function of Time



The synergistic microgel complex enhances delivery of salicylic acid into the skin. Fluorescence spectroscopy results in Figure 4 show that the 2% salicylic acid microgel complex has higher salicylic acid fluorescence intensities at all taping points and time points compared to 2% salicylic acid alone. After 60 minutes of product application and 5 tapes, the 2% salicylic acid microgel complex composition has an average fluorescence intensity twice that of the composition with 2% salicylic acid alone. The results demonstrate that the microgel complex delivers greater amounts of salicylic acid deeper into the skin. This improved penetration can be attributed to the sebum solubilizing ingredients and higher concentration of salicylic acid in the microgel.

IN VITRO ANTIMICROBIAL EFFICACY AGAINST PROPIONIBACTERIUM ACNES

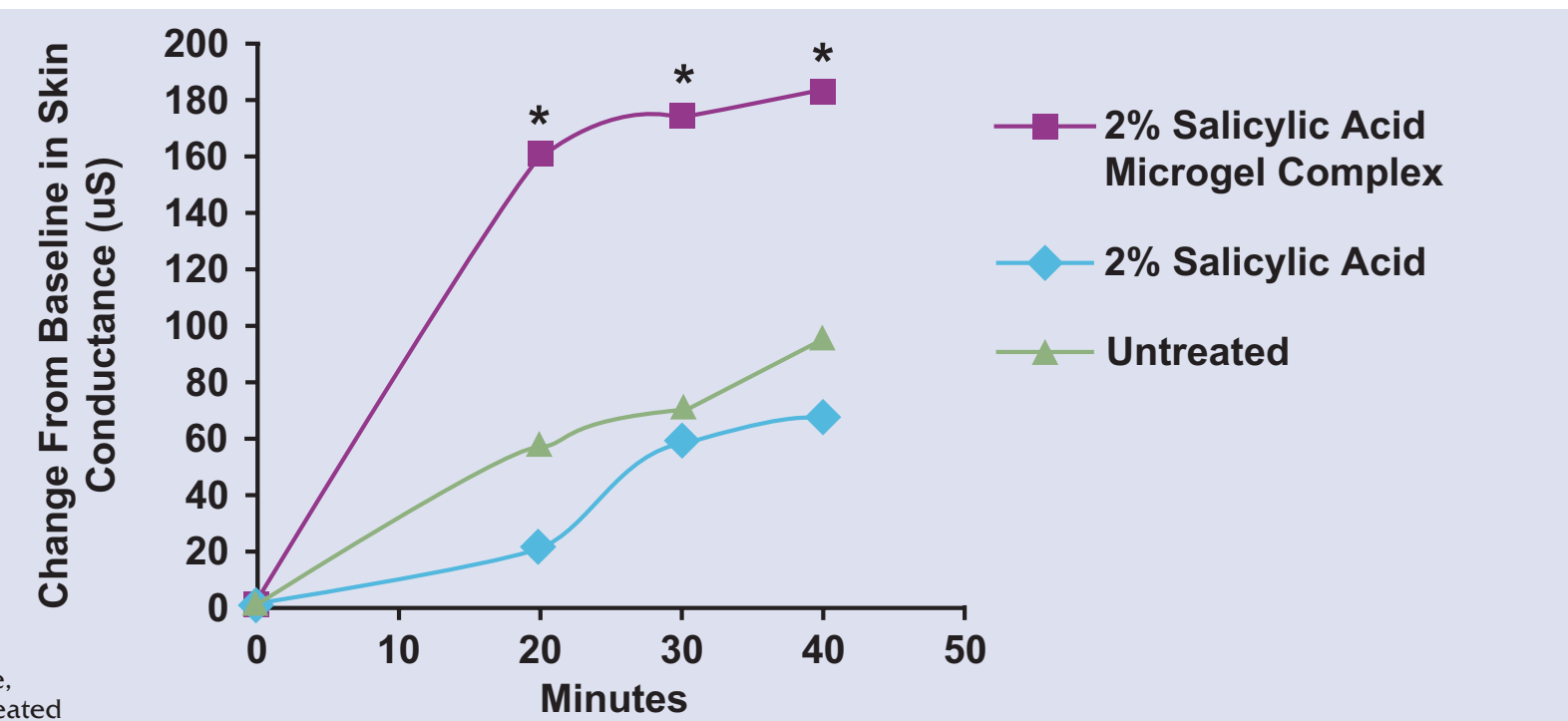
Table 1 - Log Reduction in P. acnes Population After Contact with Acne Treatment Compositions

	Log Reduction in <i>P. acnes</i> after Indicated Contact Time					
	0 Sec	15 Sec	30 Sec	1 Min	2 Min	5 Min
10% Benzoyl Peroxide Cream	0	1.3	1.5	2.6	3.1	3.1
Gel containing Novel Microgel Complex with 2% Salicylic Acid	0	3.5	3.6	3.6	3.7	3.7
Cream containing Novel Microgel Complex with 2% Salicylic Acid	0	3.5	3.6	3.6	3.6	3.6
Cleanser containing Novel Microgel Complex and 2% Salicylic Acid	0	3.5	3.6	3.6	3.6	3.6
Placebo (no microgel complex, no salicylic acid, no BPO)	0	0.53	0.67	0.93	0.82	0.88

The highlighted areas in Table 1 show that compositions with the novel microgel complex with 2% salicylic acid have greater than 3 log reduction (99.9% reduction in *P. acnes* population) starting at 15 seconds. The 10% BPO composition does not act as quickly, taking 8 times longer to reach a 3 log reduction in *P. acnes* population. The placebo composition does not show any antimicrobial activity.

SKIN BARRIER PROPERTIES IN HUMAN SUBJECTS

Figure 5 - Effects of the 2% Salicylic Acid Microgel Complex on Skin Conductance



*p<0.01 vs baseline, placebo, and untreated

Skin conductance is used to determine the moisture content of the skin.³ The skin conductance results in Figure 5 demonstrate that skin treated with the 2% salicylic acid microgel complex has a significantly greater increase in skin conductance levels versus untreated and placebo.

Skin penetration enhancers often compromise skin's barrier function, which can be measured by TEWL.³ Results show that there is no significant change in TEWL after product application; therefore, the 2% salicylic acid microgel complex does not damage the barrier function of the skin. The skin conductance and TEWL results demonstrate that the microgel complex is mild to the skin and does not dry the skin or cause barrier damage.

OXIDATIVE STRESS

Figure 6a - Effects of Topical Acne Treatments on Reactive Oxygen Species ex vivo

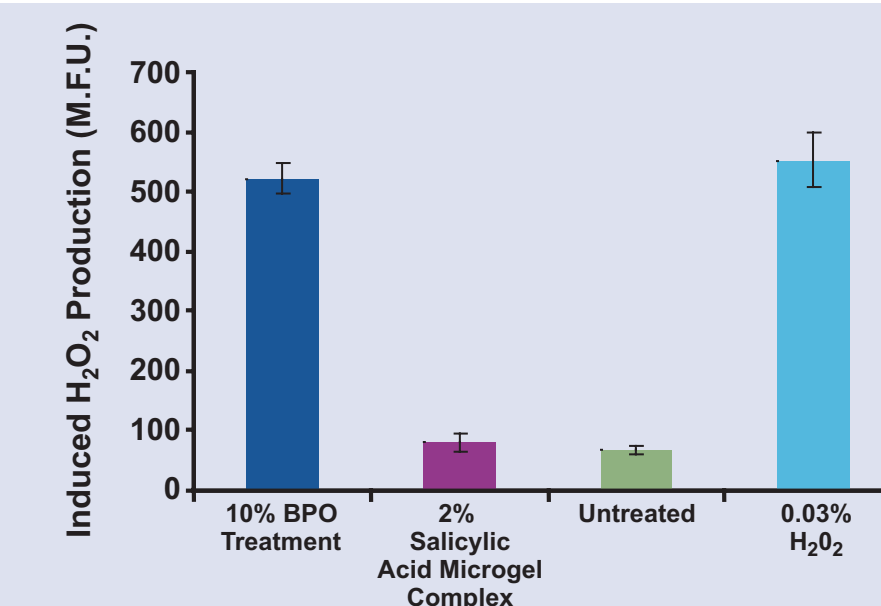


Figure 6a shows *ex vivo* levels of ROS produced by the test products measured via levels of hydrogen peroxide production. Results show that 10% BPO had a marked increase on ROS on the skin, comparable to 0.03% H₂O₂. In contrast, the 2% salicylic acid microgel complex showed no change in ROS production on the skin.

Figure 6b - Effects of Topical Acne Treatments on Reactive Oxygen Species in vivo

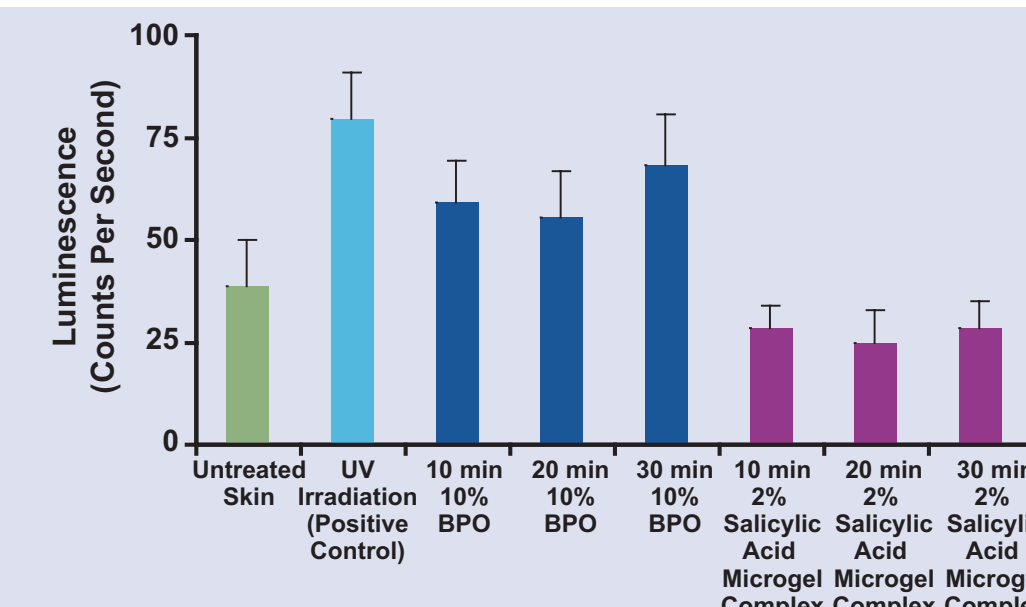


Figure 6b shows the time course of ROS production in human subjects using real time measurements of ROS-induced chemiluminescence. Results show that 10% BPO had a time dependent increase on ROS on the skin, comparable to the positive control, UVA irradiation. In contrast, the 2% salicylic acid microgel complex showed no change in ROS production on the skin.

Both *ex vivo* and *in vivo* results demonstrate the 2% salicylic acid microgel complex does not induce oxidative stress on the skin. On the other hand, 10% BPO induces significant amounts of oxidative stress. While the effects of oxidative stress are not immediately apparent to the patient, its cumulative effects can result in accelerated aging of the skin.⁴ This is an important consideration for acne treatment, especially for adult acne patients.

CONCLUSIONS

The novel microgel complex with 2% salicylic acid provides holistic acne treatment. The characterization of the novel microgel complex demonstrates the unique mechanisms of action, delivery and skin protective properties of the technology:

- Unique sebum solubilization, improved skin penetration and salicylic acid delivery
- The 2% salicylic acid microgel complex does not induce oxidative stress on the skin, while 10% BPO induces significant amounts of oxidative stress, which can result in accelerated aging of the skin
- Anti-*P. acnes* efficacy, with 99.9% reduction in *P. acnes* starting at 15 seconds
- Improves skin moisture content and does not compromise skin barrier function

The properties of the synergistic microgel complex improve patient compliance and clinical outcomes, as shown through multiple acne clinical studies.

REFERENCES

- Stamatias GN, Wu J, Kollas N. Non-invasive method for quantitative evaluation of exogenous compound deposition on skin. *J Invest Dermatol*. 2002 Feb; 118(2):295-302.
- Ou-Yang H, et al. A chemiluminescence study of UVA-induced oxidative stress in human skin in vivo. *J Invest Dermatol*. 2004;122:1020-1029.
- Wilson D, Berardesca E, Malbach HL. In vivo transepidermal water loss and skin surface hydration in assessment of moisturization and soap effects. *Contact Dermatitis*. 1988 June;201-211.
- Kevin J, Trouba, Hisham K, Hamadeh, Rupesh P, Amin, Dorl R, Germolec. Oxidative Stress and Its Role in Skin Disease. *Antioxidants & Redox Signaling*. Aug 2002, Vol. 4, No. 4:665-673.

PATENTS

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