



 **PhytoCellTec™**

## PhytoCellTec™ Alp Rose

Charges skin stem cell resistance





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## Charges skin stem cell resistance

### **Stem Cells from an Alpine Flower to Enhance the Skin Barrier**

PhytoCellTec™ Alp Rose is a powder based on the stem cells of alpine rose leaves.

The alpine rose grows at very high altitudes. Its leaves have adapted to cold, dryness and UV, so that it can resist these harsh environmental conditions and fast changes.

The skin protects our body from the environment. To perform this barrier function efficiently, the epidermis is constantly renewed and repaired by the epidermal stem cells. But environmental changes and aging impair the skin barrier leading to a drier, more fragile skin and accelerated aging. Thus, the first strategy to enhance the skin barrier is to:

- preserve the activity of epidermal stem cells
- protect them from environmental stress.

Stem cells from alpine rose leaves were obtained using our PhytoCellTec™ technology and were then incorporated in a water-soluble powder. These Alp Rose stem cells contain special epigenetic factors and metabolites which are able to preserve the human skin stem cells' function.

Using a novel Progenitor Cell Targeting technology, PhytoCellTec™ Alp Rose was shown to:

- increase the vitality of epidermal stem cells by helping them to maintain their stem cell characteristics
- protect these precious cells against environmental stress.

By optimizing the potential of epidermal stem cells, PhytoCellTec™ Alp Rose improves epidermal regeneration and thus the quality of the skin barrier.

A clinical study performed in the Alps during winter confirmed the capacity of PhytoCellTec™ Alp Rose to reinforce the skin barrier and to reduce aging signs (wrinkles, redness, loss of radiance).

### **Claim Ideas for PhytoCellTec™ Alp Rose**

- Increases skin stem cell vitality
- Boosts epidermal regeneration
- Improves the skin barrier function
- Helps the skin to cope with climate changes

### **Applications**

- Advanced "stem cell cosmetic" formulas
- Face and body care to protect the most valuable skin cells, the stem cells, against environmental stress
- Every weather formulations

### **Formulating with PhytoCellTec™ Alp Rose**

- Recommended use level: 0.4–1 %
- Incorporation: Dissolve PhytoCellTec™ Alp Rose into the aqueous phase or add pre-solved, during the cooling phase (<60 °C). PhytoCellTec™ Alp Rose is dissolvable at up to 20% in water.
- Thermostability: Temperatures of up to 60 °C for a short time do not affect the stability of PhytoCellTec™ Alp Rose.

### **INCI/CTFA-Declaration**

Rhododendron Ferrugineum Leaf Cell Culture Extract (and) Isomalt (and) Lecithin (and) Sodium Benzoate (and) Lactic Acid (and) Aqua/Water

### **Additional Information**

- PhytoCellTec™ Alp Rose contains 50% of Alp Rose stem cell extract
- Phenoxyethanol-free



# Rhododendron Ferrugineum

## Adapted to extreme environmental conditions

### A Highly Resistant Alpine Flower

PhytoCellTec™ Alp Rose is based on the stem cells of alpine rose leaves.

The alpine rose is an evergreen plant which grows at high altitudes (2000 m) in the Alps but also in the Jura, Pyrenees and Apennines in acidic and nutrient-poor soils. It has dense and extensive roots and spectacular bell-shaped flowers. Its leaves have adapted to cold and dryness:

- they are coated by a thick cuticle
- on their undersides, scales cover stomates and thus limit water loss.

The rust-brown color of these scales gives this species its scientific name, *Rhododendron ferrugineum* ("rhodon" for rose, "dendron" for tree and "ferrugo" for rust).

### Adapted to Extreme Environmental Variations

The alpine rose can live for more than 100 years. This cold-resistant shrub is able to adapt to very hard environmental conditions such as large variations in temperature, UV and dryness. Its leaves, which are well protected by snow during winter, are suddenly exposed to intense sunlight and temperature when the snow melts. High contents in various polyphenolic compounds protect the plant tissue against attack of radicals.

In response to cold, dehydrating or freezing stresses, alpine rose leaf tissues start to synthesize protective proteins called dehydrins. Dehydrins accumulate during cold acclimatization and behave as tiny sponges by retaining water around cell membrane and proteins.

### Used to Cope with Weather Changes

Today, *Rhododendron ferrugineum* is mainly used in homeopathy to treat neuralgia and rheumatism symptoms aggravated by weather change (thunderstorm, wind, humid cold).

### Threatened by Climate Change

Global warming accelerates snow melt. This has a negative impact on the alpine rose which is protected by snow during winter.

In order to preserve biodiversity and prevent its extinction, the alpine rose has been registered in the Swiss database for the conservation and sustainable use of aromatic and medicinal plants for food and agriculture. In some cantons in Switzerland, it is forbidden to pick it.

# PhytoCellTec™ Alp Rose

## Environmental stress impair the skin barrier

### Skin Barrier is Controlled by Epidermal Stem Cells

The epidermis and notably the stratum corneum, its upper layer, protects the organism from the environment and external stress while simultaneously preventing water loss.

To perform this barrier function while confronting the environment, the epidermis undergoes constant maintenance, renewal and repair. This process is ensured by essential cells, the epidermal stem cells which are located in the basal layer, the deepest layer of the epidermis. There, epidermal stem cells replenish the epidermis with new cells. When keratinocytes migrate from the basal layer, they no longer divide but they undergo differentiation to form the outermost horny layer (stratum corneum).

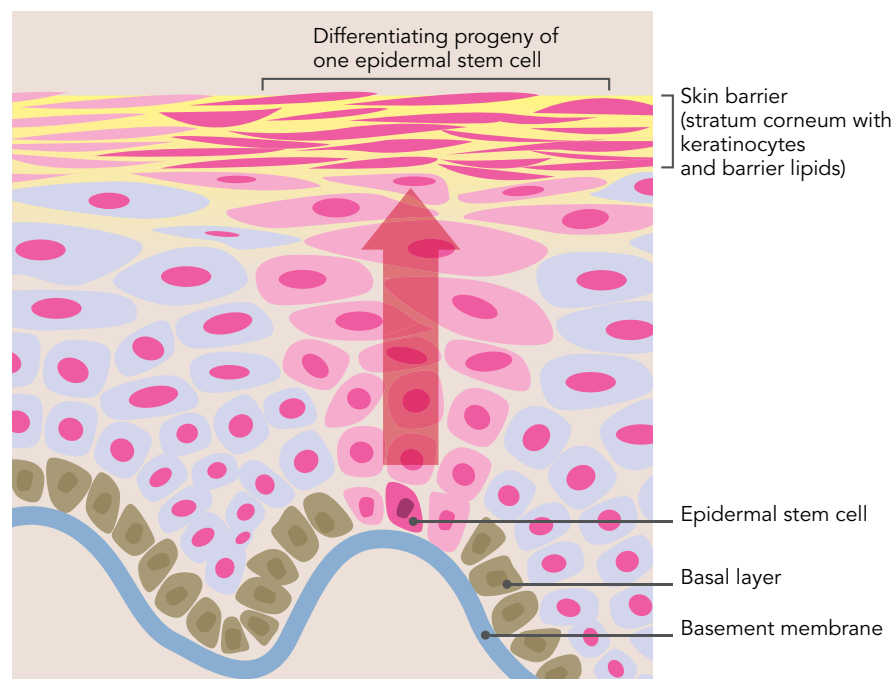
### Environment and Aging Compromise the Skin Barrier

Low temperatures, low humidity and abrupt environmental changes impair the skin barrier.

With age, the skin renewal is slowed down and the production of key barrier lipids is reduced. Besides, stem cells from older donors have been found to be less effective than those from younger ones.

Thus, both environmental factors and aging compromise the barrier function. Less protected, the skin becomes more susceptible to environmental stress. This results in a drier skin and accelerated aging.

### Role of Epidermal Stem Cells in the Skin Barrier Function



# PhytoCellTec™ Alp Rose

## Alp Rose stem cells to protect epidermal stem cells and to preserve the skin barrier

### Plant Stem Cells for Skin Stem Cells

Because epidermal stem cells control the formation, maintenance, renewal and repair of the whole epidermis, the first strategy to enhance the skin barrier is to:

- preserve the function of epidermal stem cells
- protect them from environmental stress.

All stem cells, independent of their origin (plant, animal or human) contain specific epigenetic factors whose function is to maintain the self-renewal capacity of stem cells. Therefore the Alp Rose stem cells are used to help to preserve the vitality of human skin stem cells.

### Protective Compounds to Support Skin Barrier

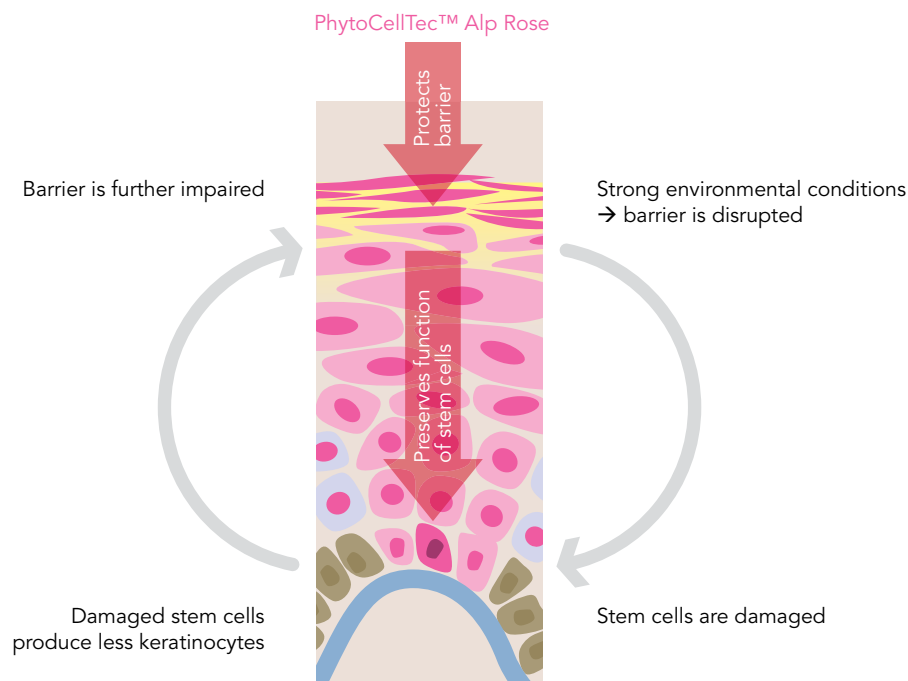
At its extreme mountain habitat the alpine rose had to adapt to very hard environmental conditions such as exposure to intense UV light and big temperature differences. The plant is therefore especially rich in polyphenolic compounds with antioxidant and radical scavenging properties.

Well documented is the presence of hyperoside, myricetin, kaempferol, quercetin and taxifolin. Characteristic for alpine rose is the phenolglycoside rhododendron that was shown to exert anti-inflammatory effects.

The winter period is a special challenge, not only because of minus temperatures but also because of winter dryness. If the water in the stem is frozen then it cannot be mobilized anymore for the plant tissues. In order to prevent damage from dehydration stress, the cells of the alpine rose start to synthesize special protection proteins, called dehydrins. They are very rich in glycine residues and are known to accumulate during dehydration stress caused by freezing temperatures, drought or salinity.

In a chaperone-like manner, dehydrins stabilize other proteins and membranes by coating them with a cohesive water layer. In the same way, these cover proteins will impart benefits to our skin barrier.

### Mechanism of PhytoCellTec™ Alp Rose



# PhytoCellTec™ Alp Rose

## Advanced biotechnology to cultivate plant stem cells

### PhytoCellTec™ by Mibelle Biochemistry

Mibelle Biochemistry has developed a novel technology called PhytoCellTec™, enabling the large scale cultivation of cells from rare and endangered plant species or from plants difficult to harvest.

This technology is based on the unique totipotency of plant cells that is to say:

- the capacity of every plant cell to regenerate new organs or even the whole plant
- the capacity of every plant cell to dedifferentiate and become a stem cell.

Our PhytoCellTec™ technology relies on the wound healing mechanism of a plant: part of a plant is wounded to induce the formation of callus cells. This healing tissue consists of dedifferentiated cells which are stem cells. Callus cells are harvested and cultivated in suspension. A novel bioreactor system enables the large scale production. Alp Rose stem cells are then submitted to low temperature in order to acclimatize them to cold.

To obtain the PhytoCellTec™ Alp Rose cosmetic ingredient, these stem cells are harvested and homogenized at 1200 bar together with phospholipids to encapsulate and stabilize oil- and water-soluble components into liposomes. The resulting extract is carefully sprayed on a powder based on isomalt.

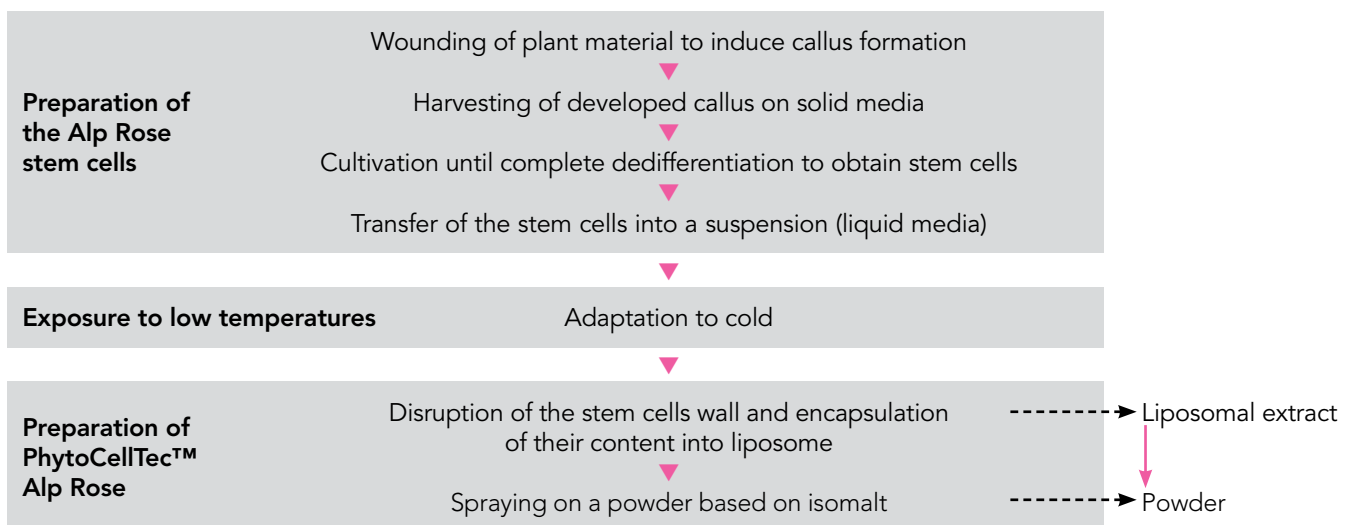
PhytoCellTec™ Alp Rose is thus rich in epigenetic factors and metabolites which preserve the function of skin stem cells and protect them against environmental stress.

### Advantages of PhytoCellTec™ Technology

This innovative technology developed by Mibelle Biochemistry offers the following advantages:

- possibility to cultivate cells of rare and endangered plants based on small amounts of origin plant material
- availability of plant material independent of the season and market demand
- plant material completely free of environmental pollutants and pesticides
- constant concentrations of metabolites in the stem cells.

### PhytoCellTec™ Alp Rose Process



# PhytoCellTec™ Alp Rose

## Study results



### Preparation of Epidermal Stem Cells

A novel Progenitor Cell Targeting technology was used to prepare human epidermal stem cells.

It consists of culturing a skin sample in a medium specifically designed to mimic the micro-environment of the stem cell in the epidermis. This leads to an enrichment of so called keratinocyte progenitor cells that can be considered as stem cells. This enrichment (10-fold) was quantitatively controlled through FACS (Fluorescence-activated cell sorting) of cells which were labelled with CD34 and  $\alpha 6$  integrin, two well-known markers of epidermal stem cells. This technology enables us to specifically evaluate the effect of a compound on epidermal stem cells.

Stem cells have the characteristic to form colonies in vitro. A colony consists of the progenitor cell, transient amplifying cells (cells in an intermediate state) and differentiated cells that have lost the capacity to divide. The number of colonies formed is a value of the vital progenitor/stem cells and is called colony forming efficiency (CFE).



### Maintenance and Protection of Epidermal Stem Cell Function

The epidermal stem cells obtained using the Progenitor Cell Targeting technology, were treated with and without the Alp Rose stem cell extract. Some were then exposed to UVA+UVB-light at 450 kJ/m<sup>2</sup> whilst others were left unexposed. Afterwards, the CFE was determined.

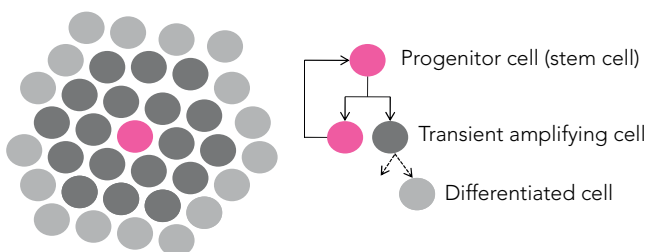
Results showed that:

- the CFE was increased by 75% in the presence of 0.15% Alp Rose stem cell extract
- the Alp Rose stem cell extract was able to maintain the CFE even when epidermal stem cells were exposed to UV radiation.

This clearly shows that Alp Rose stem cell extract:

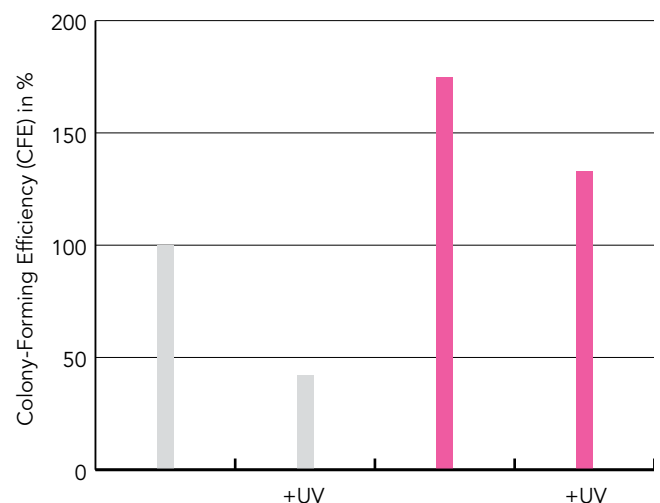
- increases the vitality of epidermal stem cells by helping them to maintain their stem cell characteristics ("stemness")
- protects epidermal stem cells against UV-induced stress.

### Capacity to Form a Colony (CFE)



### Improvement of the Vitality of Skin Stem Cells

■ Control ■ Alp Rose stem cell extract 0.15%







### Protection of the Skin against Harsh Environmental Conditions

In a study performed with 22 Caucasian volunteers of phototype II and aged from 20 to 52 (mean = 41.3 years old) during ski holidays, PhytoCellTec™ Alp Rose was shown to protect the skin barrier.

Volunteers applied 3 times a day (morning, noon and evening):

- a sun cream SPF 30 (placebo) on one half of their faces
- the same sun cream SPF30 enriched with 0.4% PhytoCellTec™ Alp Rose on the other half of their faces.

The application of test products started 2 days before the ski holidays week and continued 1 week after, corresponding to 16 days use.

The ski holidays took place in the Alps, in the first week of March 2010.



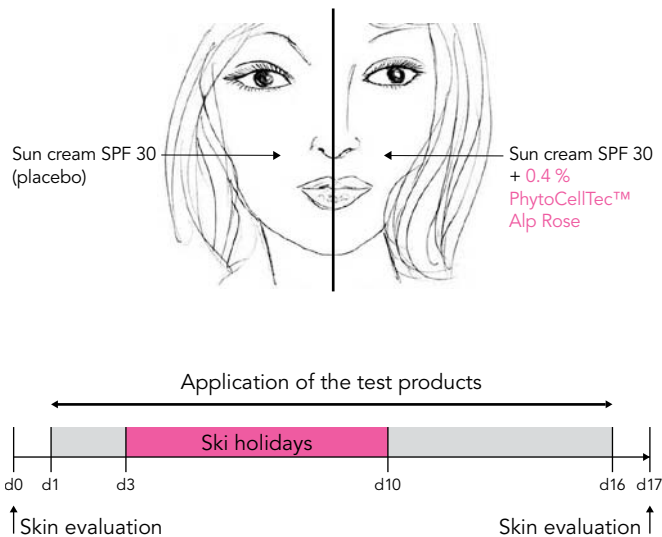
### Protection of the Skin Barrier

After the use of PhytoCellTec™ Alp Rose for 16 days, the TEWL was shown to be reduced compared to the placebo.

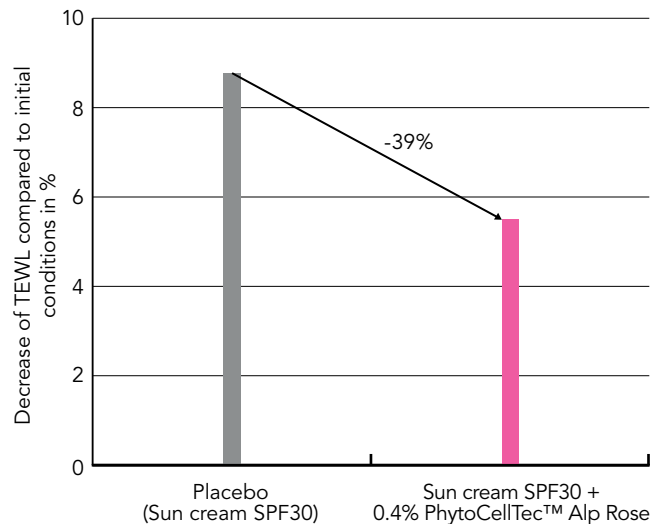
The TEWL was measured on the face (cheeks) using a tewameter.

This 39% reduction of TEWL indicates a stronger skin barrier. Thus, PhytoCellTec™ Alp Rose protects the skin barrier by making it more resistant to the combination of strong environmental stresses (UV, cold, wind, dryness).

## Study Design



## Reduction of TEWL on the Face



# PhytoCellTec™ Alp Rose

## Study results



### Anti-Wrinkle Effect

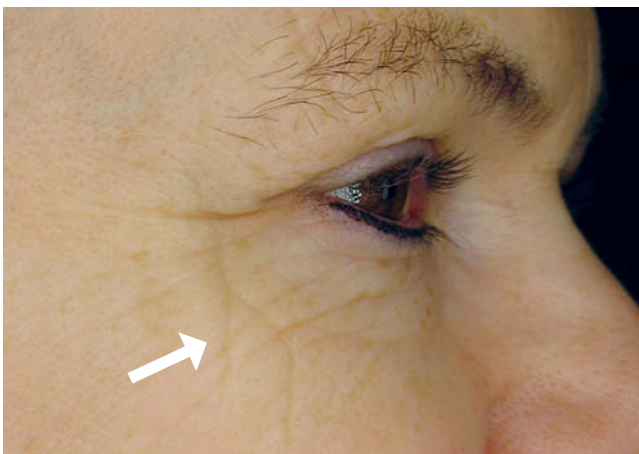
During the previously described study, high-definition photographs were taken using the VisioFace Quick®.

On these photographs taken with white light, the characteristics of the skin such as wrinkles, skin redness and skin radiance were evaluated by trained assessors.

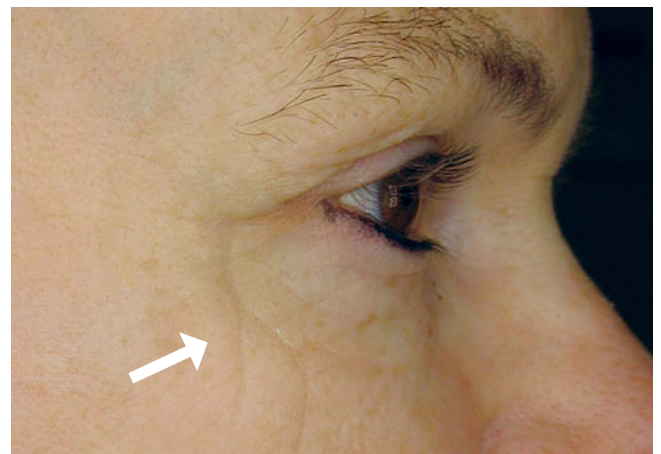
Results showed, after only 16 days a visible reduction of the depth and number of wrinkles on crow's feet by PhytoCellTec™ Alp Rose.

### Anti-Wrinkle Effect on Crow's Feet after 16 Days of Use

Before (day 0)



After (day 17) 0.4% PhytoCellTec™ Alp Rose





### Visible Improvement of the Skin Quality

Before (day 0) and at the end of the study (day 17), the following characteristics of the skin of all volunteers were scored visually by trained assessors:

- wrinkles
- radiance of the complexion
- redness / irritations

On the half side of the face treated with the placebo, results didn't show any improvement of the skin radiance and redness / irritations compared to initial conditions and even a worsening of the wrinkles.

Comparatively, the other side treated with PhytoCellTec™ Alp Rose showed an improvement of each of these 3 characteristics.

Compared to initial conditions and to the placebo:

- wrinkles were less visible for 45% of the volunteers
- complexion was more radiant for 54% of the volunteers
- redness/irritations were reduced for 9% of the volunteers.



### Perception of the Efficacy by Self-Evaluation

At the end of the study, the volunteers were asked, using a questionnaire, to tell if they perceive a general better skin feeling on one side of their face, and if yes, on which side.

Criteria for a better skin feeling were:

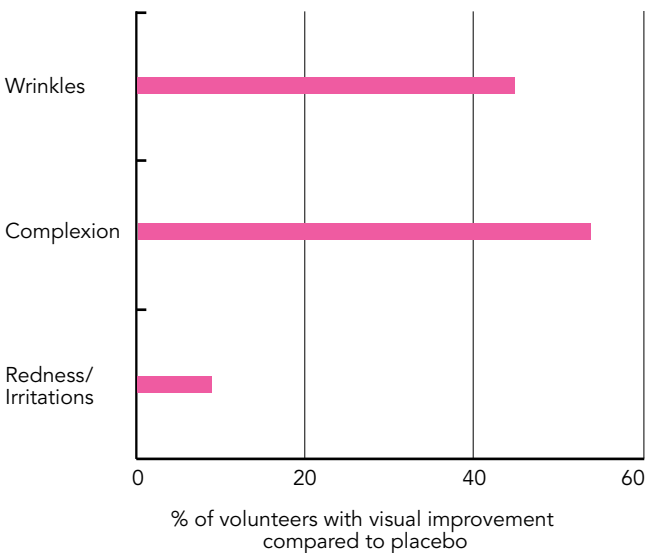
- overall general improvement
- better protected skin
- less easily irritated skin
- skin better protected from cold

Results showed that:

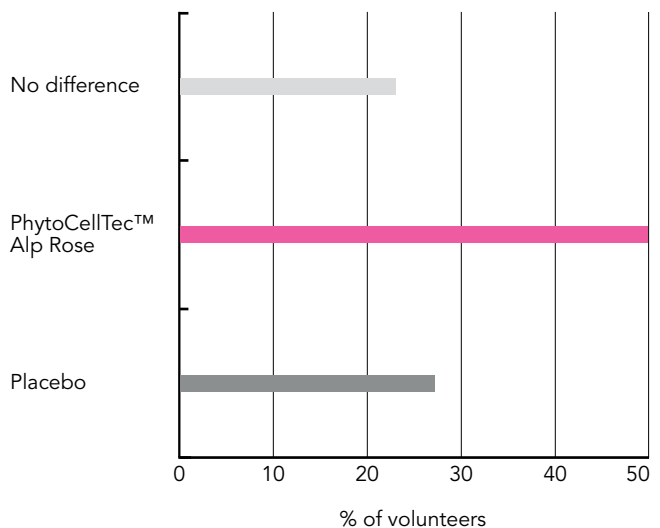
- 22.7% of volunteers didn't perceive any difference between the two halves of their faces.
- 27.3% of volunteers perceived an improvement on the side treated with the placebo.
- 50% of volunteers perceived an improvement on the side treated with PhytoCellTec™ Alp Rose.

## Improvement of Skin Quality

■ PhytoCellTec™ Alp Rose



## Perception of a General Improvement



# PhytoCellTec™ Alp Rose

## Charges skin stem cell resistance

### Claim Ideas for PhytoCellTec™ Alp Rose

- Increases skin stem cell vitality
- Boosts epidermal regeneration
- Improves the skin barrier function
- Helps the skin to cope with climate changes

### Applications

- Advanced "stem cell cosmetic" formulas
- Face and body care to protect the most valuable skin cells, the stem cells, against environmental stress
- Every weather formulations

### Marketing Benefits

- Plant stem cells to protect skin stem cells (patent pending)
- Proven efficacy on skin stem cells
- PhytoCellTec™ technology – recognized as an "eco-breakthrough" at Rio+20, the conference of the United Nations on sustainable development
- Tested in harsh environmental conditions
- Visible anti-aging effect after only 16 days
- [www.phytozelltec.ch](http://www.phytozelltec.ch)



### Innovating for your success

Mibelle Biochemistry designs and develops innovative, high-quality actives based on naturally derived compounds and profound scientific know-how. Inspired by nature – Realized by science.

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