



Provital  
Do Care



# KERAMARE™

CareActives

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Shine



## INTRODUCTION

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Day after day we hear that the first impression, we get of a person is the one that matters, because in just a few seconds we can create an idea about his or her personality and character. In these first moments, a perception is created in the other person based on his or her gestures, the way he or she communicates, his or her outfit, or other factors, such as hairstyle or makeup.

Nowadays, we have tools to enhance our image and make a positive impact on others. One factor that can be easily modeled is hair, a very important element of our appearance.

In addition, hair appearance not only influences other people, but it also has an influence on ourselves. It is important to have healthy, fresh, free-moving hair, as it can directly influence character and mood, and it can define professional, personal and even emotional success. To demonstrate the influence of hair on a person, in 2009 a study was conducted in which it was considered that 56% of European women feel more attractive and confident when their hair shines by itself. Every day, an increasing number of women and men undergo treatments to enhance the appearance of their hair. These treatments can be **chemical** treatments, such as dyes, or **thermal** treatments, such as the use of hair straighteners and hair dryers. Moreover, we are exposed to other factors on a daily basis, such as exposure to different **environmental aggressions** (UV, wind, extreme temperatures, etc.). All these factors can damage hair from the **inside**, causing a loss of fiber integrity and structure, towards the **exterior**, leading to a rough, weak, limp and dull appearance.

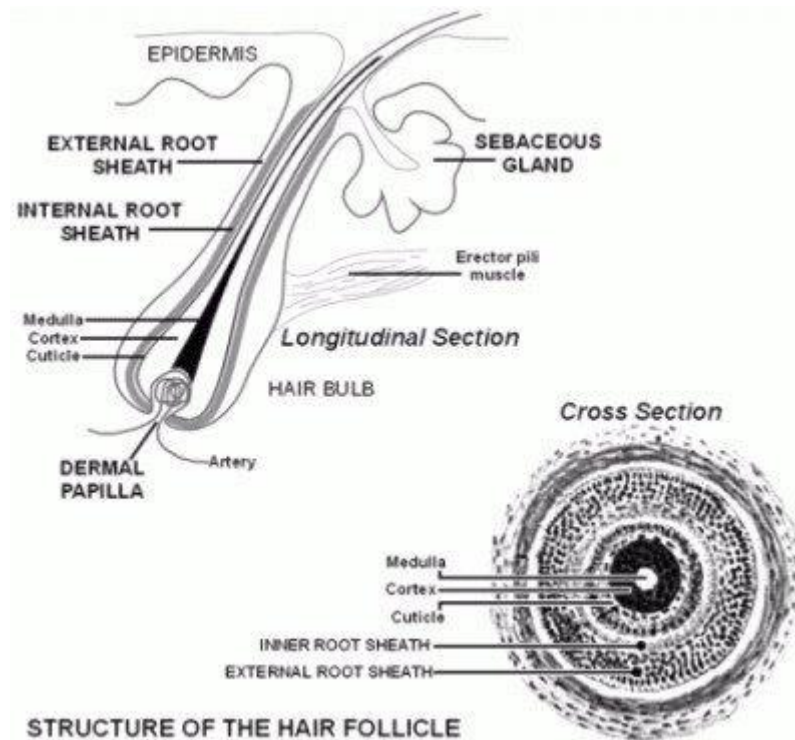
**KERAMARE™ is an active ingredient specifically designed to repair and restore the natural conditions of healthy hair. It works from the inside to restore the integrity of hair fibers, and its action is reflected on the outside, improving visual and sensory parameters such as appearance, moisturization, touch and shine.**



## HAIR MORPHOLOGY

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Hair is a set of cylindrical and flexible filaments that are arranged obliquely in relation to the surface of the skin. Two parts can be differentiated: an internal part called **hair follicle** and the external part known as **stem**.



### Hair follicle

Each hair has a hair follicle, from which it emerges. The hair root is located inside this hair follicle and it arises from a round expansion called hair bulb, which is the proliferative area of the follicle.

Around the follicle there is the basal lamina, the pilo erector muscle, a sebaceous gland that lubricates and protects hair from dehydration, and vascular and nerve endings that reach the dermal papilla.

### Shaft

The shaft is the visible part of hair, a long cylinder composed of keratinized cells. Three layers can be differentiated from the inside towards the outside: *medulla*, *cortex* and *cuticle*.



- ✓ **Medulla:** It appears sparsely and even intermittently in the hair shaft. Its function has not been specified. It contains small enucleated cells with small amounts of lipids.

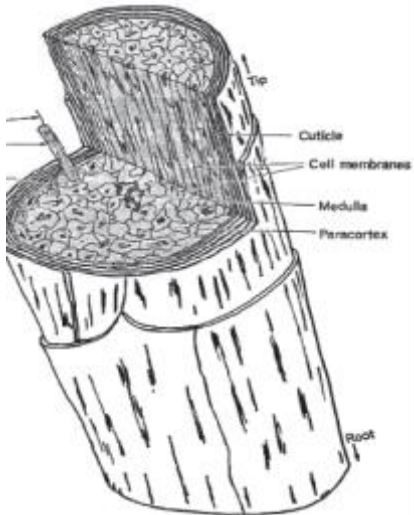


Figure 2. Cutaway view of a hair fiber

- ✓ **Cortex:** It is the central part of the shaft, the strongest and the most important one. Hair diameter depends on this layer which, in turn, is proportional to the number of cells that are undergoing mitosis in the bulb. Hair resistance depends on its condition. Cortex cells are formed by an amorphous protein matrix at 40%, with a high sulfur proportion, and by a fibrillar protein at 60%, with a small amount of sulfur and a helical shape. Two areas can be differentiated in the cortex:

- *Ortho-cortex:* with a higher density of fibrils and a lower proportion of amorphous protein matrix.
- *Paracortex:* with fewer fibrils and more matrix.

- ✓ **Cuticle:** It protects the internal structure, as it covers the hair shaft. A

**healthy, shiny and smooth** appearance depends on this layer. As they ascend along the follicle, precursor cells differentiate and flatten into layers, called scales. They tilt up and overlap each other resulting in a structure similar to shingles on a roof. They contain keratin with a harder consistency than that of epidermal cells.

## HAIR KERATIN

Hair consists mainly of proteins, water, lipids, pigments and other components. The main protein is **keratin**, which constitutes 80% of human hair (Robles Velasco et al., 2009). It consists of polypeptide chains that are arranged in a helical shape parallel to a longitudinal axis.

Keratins are proteins insoluble in water, which can adopt two conformations:  **$\alpha$ -helix or  $\beta$ -sheet keratin.**

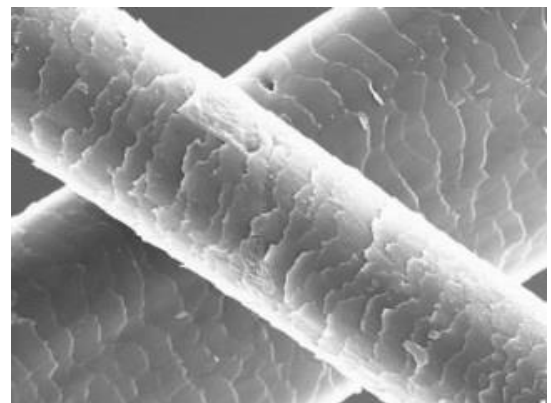


Figure 3. Microscopic image of a hair fiber

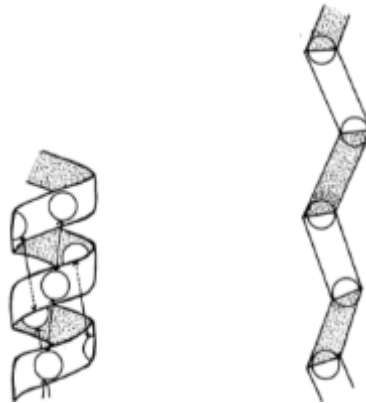


Figure 4.  $\alpha$ -helix and  $\beta$ -sheet keratin

Most of the hair is composed of **keratin with an  $\alpha$ -helix conformation**. The polypeptide chain of this protein is wrapped in a  $\alpha$ -helix, and several of these helices form a **protofibril** (=filaments). Eleven of the protofibrils combine and form aggregates called **microfibrils** (cell intermediate filaments). In turn, hundreds of these combine to form a protein structure called **macrofibril**. Therefore, hair fibers are constituted by the stacking of cells formed by macrofibrils, which gives hair an extraordinary resistance (Fig. 5).

The amino acid composition of the cuticle and the cortex shows the same trend. Mostly over 70% are neutral amino acids. Acidic amino acids are around 18-11% and basic amino acids are present in a lower proportion, between 6-9%. Hair's global charge is **negative**, due to the side chains of acidic amino acids, taking into account that the isoelectric pH of keratin is 3.8 (Robles Velasco et al., 2009).

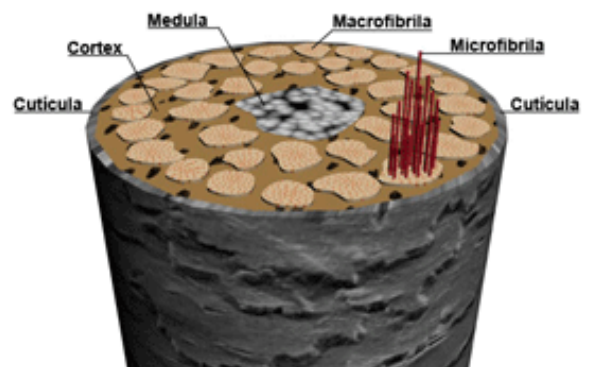


Figure 5. Hair fiber

#### COMPOSITION: BOTANY AND CHEMISTRY

**KERAMARE™** is a marine active ingredient obtained from *Cystoseira compressa* (Esper) Gerloff & Nizamuddin, which basically consists of sulfated polysaccharides. In addition, it forms a complex with a specific Matrix, which enables a targeted action on the hair fibers to assist in the fixation of the active ingredients (sulfated polysaccharides).



This specific **Matrix**, is a complex formed by amylopectin and **KERAMARE™**, and represents an improvement in the performance of hair care treatments, as it enhances their distribution, deposition and action, due to their sequential release.

### ***Cystoseira compressa*, MARINE ACTIVE INGREDIENT**

#### **Botany**



Figure 6. *Cystoseira compressa* (Esper) Gerloff & Nizamuddin

*Cystoseira* is a genus of perennial brown algae (*Phaeophyceae*) belonging to the order Fucales. It is characterized by highly differentiated basal and apical regions, and by the presence of chained pneumatocysts (air vesicles), which maintain the algae in an erect position, seemingly floating in strong currents.

Mature specimens have a main axis and proportionately elongated sides. The bottom parts are strongly flattened in “leaf expansions” or “basal leaves”. The fertile regions support conceptacles located in receptacles. These are usually located at the tips of the branches and contain the reproductive cells of algae.

*Cystoseira compressa* is a dark-brown alga that can reach up to 30 cm high. It develops both in the Mediterranean and on the two sides of the Atlantic (Caribbean, Azores, Canary Islands, Atlantic coasts of the Iberian Peninsula), in the infralittoral area with well-lit rocky bottoms, with variable hydrodynamics and bathed in clean water. It is attached to the bedrock by a relatively small basal fixation disk.

#### **Chemistry**

Brown algae, such as *Cystoseira compressa*, contain photosynthetic pigments that allow them to perform photosynthesis at different levels or depths in the ocean. These pigments are mainly chlorophyll a and c, fucoxanthin and diatoxanthin.

Their cell wall contains cellulose, alginic acid and sulfate salts, compounds that give strength and flexibility to the algae, as they form gels in the intercellular matrix that help them withstand the stresses caused by waves and sea currents. In addition, they are involved in ion exchange and help prevent algae desiccation.



Moreover, they contain **fucanes**, which are a family of polysaccharides extracted from brown algae and which consist mainly of a chain of position 3 bonded  $\alpha$ -L-fucose units, some of them sulphated and with simple L-fucose branches. When there are also units of xylose, uronic acids, galactose and mannose present, polysaccharides are called **fucoïdants** (fig.7).

Fucoïdants have a proven potential for topical or nutraceutical applications, as they can be easily incorporated into cosmetic

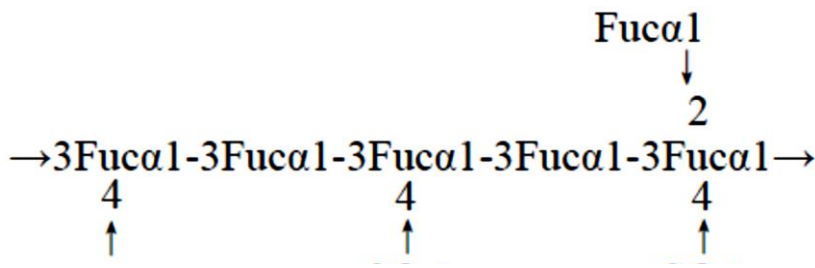


Figure 7. General structure of fucanes (Li Bo, 2008).

products, providing diverse activities such as anti-aging, anti-inflammatory (Fitton et al., 2007) or, in this case, for hair application.

#### **AMYLOPECTIN, MATRIX COMPONENT**

To maximize the efficacy of a cosmetic active ingredient on the hair, the active ingredient has to remain on the hair surface long enough to obtain a cosmetic benefit. To this end, it is necessary to have an application mechanism that enables this. **KERAMARE™** is an active ingredient that meets this need, as it presents its hair active ingredients in a controlled release tridimensional hydrocolloid matrix (Matrix).

#### **Matrix, the sequential release matrix**

This matrix consists of amylopectin, a non-ionic component that produces the Matrix, a controlled sequential release system of hair active ingredients. This matrix attaches to the hair fiber and increases contact time with the active ingredients it releases (sulfated polysaccharides in **KERAMARE™**), which will improve hair characteristics.



Amylopectin is a non-ionic water-soluble biopolymer derived from maize (*Zea mays*) composed of glucose units linearly linked by  $\alpha$  bonds (1-4). Its ramified structure gives it a configuration capable of containing between 100,000-200,000 glucose molecules and it facilitates the preservation of the product on the surface, but unlike linear polymers it prevents excessive accumulation (Hössel, 2000).

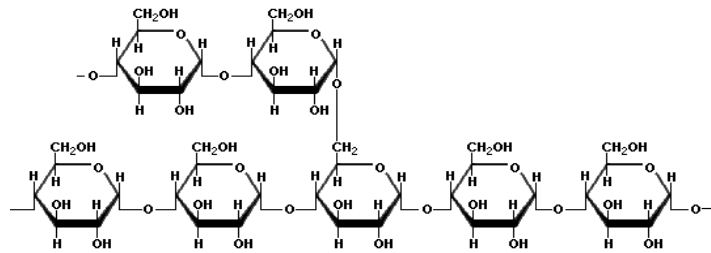


Figure 8. Amylopectin structure

#### THE MATRIX: TECHNOLOGY AND FUNCTIONALITY

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The structure formed by **the Matrix with the injected KERAMARE™**:

- ✓ Seals the active ingredients and the humidity inside the fiber using a protective film.
- ✓ Achieves a better product deposition on the hair.
- ✓ Improves the distribution of the active ingredients and prolongs their action time.

**By combining the properties of *Cystoseira compressa* and the Matrix, a spectacular improvement in the sensory and physical characteristics of hair is obtained**





## PROPERTIES: EX VIVO EFFICACY

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### MATERIALS

The hair used in this project has been dark brown natural hair subsequently bleached using a chemical treatment. The strands of hair have been treated with neutral shampoo, without active ingredient, and with two types of serum depending on the sample: one serum contains the **KERAMARE™** active ingredient at 5%, and the other one is the placebo, that is, without incorporated active ingredient.

### METHODS

#### Degradative hair treatment

Bleached hair has been obtained from 40 g of untreated hair, which has been submerged for 30 minutes, at 25°C and with stirring, in a solution consisting of 9% of H<sub>2</sub>O<sub>2</sub>. Subsequently, the hair has been washed and left to dry at room temperature. This procedure has been repeated 15 times, preparing a new solution for bleaching each time.

#### Application of the formulations on the hair

The formulations have been applied on the hair using the following experimental method. Three samples of bleached hair and one sample of untreated hair are prepared. All hair samples are washed with neutral shampoo daily for 2 minutes and then rinsed with water. Then, the formulations are applied on the hair and they are let stand for 24 hours. After this time, the samples are washed with the neutral shampoo and the formulations are reapplied. This procedure is repeated 10 times. In the end, the following hair samples are available:

- ✓ **UT:** Untreated hair
- ✓ **B:** Untreated bleached hair
- ✓ **BP:** Bleached hair + Placebo
- ✓ **BM:** Bleached hair + **KERAMARE™**



## TESTS

### 1. Sensory evaluation

Sensory analysis allows us to measure, describe, evaluate and interpret a series of hair properties using the senses as measurement instruments. Sight and touch help us appreciate highly valued cosmetic properties, such as hair appearance, brightness, touch and dryness. This type of assessment provides valuable information about the condition of hair after undergoing chemical treatments and how it is affected by common cosmetic treatments. It is common practice to determine the efficacy of hair care treatments.

Subjective assessment has been used, after the bleaching and subsequent application of the different cosmetic formulations, to find out **the overall effect on the hair**.

A trained panel has carried out the evaluation of various characteristics of the different hair samples. The scoring of these parameters may vary from value 1 (poor appearance, rough, dull, dry) to 5 (very good appearance, very smooth, very bright, highly moisturized).

### Results

The overall results of the subjective assessment of the different hair samples are shown in the chart below:

Results show that bleaching (B) produces a damaging effect on the hair, with a decrease in its appearance, touch, brightness

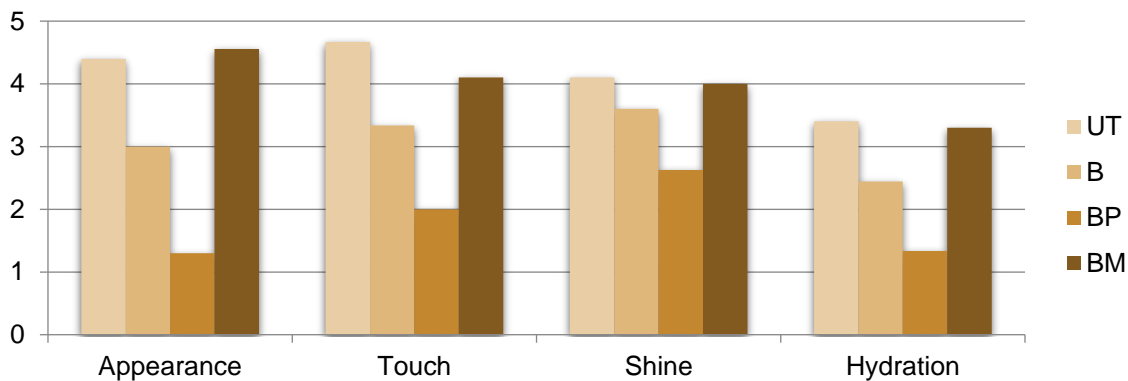


Chart 1. Mean values of the subjective assessment of appearance, touch, shine and moisturization for untreated hair (UT), bleached hair (B), bleached hair treated with placebo (BP) and **KERAMARE™** (BM).

and moisturization when compared with untreated hair (UT). It can also be observed that the bleached hair treated with placebo (BP) is equally considered as damaged, with a clear deterioration in all assessed subjective parameters compared to the rest of hair samples.



The treatment with **KERAMARE™** (BM) presents a better assessment in all subjective parameters. Results show that the natural appearance of hair is improved, as the harmful effects of the chemical treatment are repaired.

Figures 9 and 10 are an example of the hair samples obtained with the different treatments:



Figures 9 and 10. Samples of untreated hair (UT), bleached hair (B), bleached hair treated with placebo (BP) and bleached hair treated with **KERAMARE™** (BM).

**KERAMARE™ recovers and improves hair's natural appearance,  
as it repairs the damage occurred**

## 2. Brightness

Brightness is one of the most valued hair properties, as it implies hair health and beauty. It measures the ability of surfaces to reflect light without scattering it. This characteristic is closely linked to the structure of the hair fiber and, in particular, it depends on the condition of the cuticle.

Over time, hair is exposed to many environmental processes and agents that mainly damage the outermost layer of hair, that is, the cuticle. In consequence, hair is stripped of its natural protection, and it gradually loses its original brightness and smoothness, characteristics related to healthy hair (Schueller et al., 2001).

The assessment methods of hair brightness comprise instrumental methods (Keis et al., 2004) and subjective methods. The ideal situation for any product is to perform a combination of instrumental methods, which are objective, together with

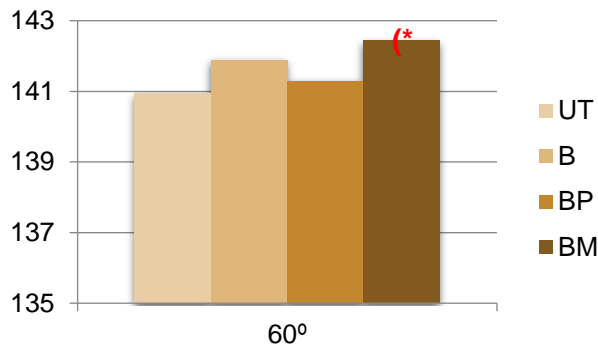


other subjective tests, where consumers explain the perception they observe after the application of the hair cosmetic product.

An objective assessment of the brightness can be performed using the glossmeter. Hair samples are combed to arrange them in parallel and measurements are carried out through a microscope slide placed on the hair. This process is used to visualize **the condition of the hair cuticle** after bleaching and the effect caused due to the application of the various cosmetic formulations.

### Results

Table 1 shows the average values obtained in the brightness assessment using an angle of incidence of 60°. Results have been statistically evaluated to estimate whether the differences in brightness are significant.



The variations in brightness values at 60° in the fibers treated

Table 1. Brightness average value (in brightness units) of hair at 60° for untreated hair (UT) and bleached hair (B), and for bleached hair treated with the placebo (BP), and hair treated with **KERAMARE™** (BM) (\*p<0.05).

with **KERAMARE™** show a significant increase in brightness compared to untreated samples (UT) and samples treated with the placebo (BP).

**KERAMARE™ repairs the dull and damaged appearance of hair, so it shines with its own light**



### 3. Increase in hair resistance and thermal protection by using differential scanning calorimetry (DSC) technique

Differential scanning calorimetry (DSC) is a technique that allows us to know the organization and stability of the three-dimensional structure of proteins, such as keratin. DSC measurement has been used to determine the **integrity of the  $\alpha$ -helix structure in the cortex** after bleaching, and the effect caused due to the application of the various cosmetic formulations (Wortmann et al., 1998).

Differential scanning calorimetry testing, performed in dry conditions, allows us to thermally characterize keratin fibers through the observation of four thermal processes: vitreous transition, water evaporation, protein denaturation and thermal decomposition or pyrolysis.

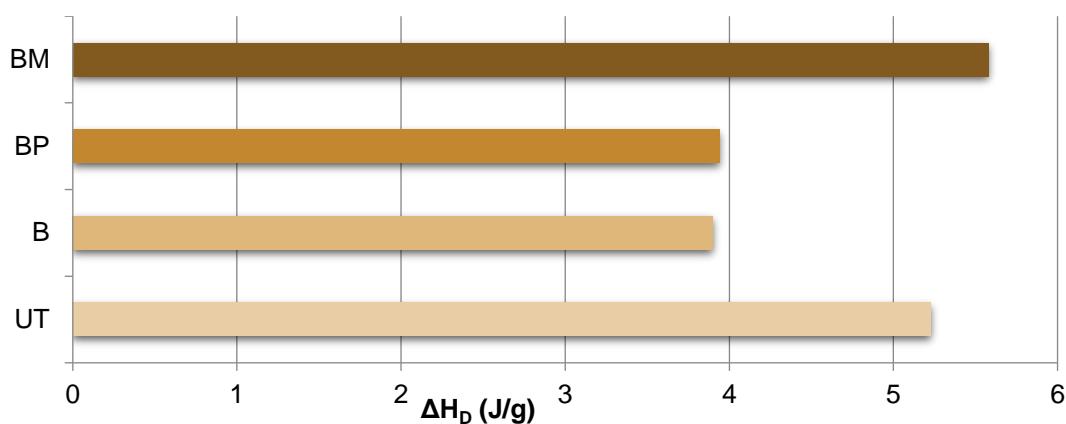
Protein denaturation occurs between 230 and 250°C, where two peaks are observed representing structure loss in two hair parts: the **ortho-cortex** and the **paracortex**. Wortmann and Deutz (1998) observed this dual behavior of the cortex when they conducted a thermotropic study of the fibers, demonstrating that the denaturation temperature of the paracortex was higher than that of the ortho-cortex.

#### Results

The peak corresponding to protein denaturation was characterized by its enthalpy ( $\Delta H_D$ ).

Bleaching produces an appreciable decrease in the enthalpy of the denaturation peak ( $\Delta H_D$ ), as it damages the crystalline material of hair.

If we take a look at the results for the ortho-cortex (chart 2), the use of **KERAMARE™** rises the increase in enthalpy ( $\Delta H_D$ ),



and this demonstrates that this active ingredient exerts a **restructuring action of keratin fibers**.

Chart 2. Enthalpy variation in the ortho cortex.



The ortho-cortex contains a higher density of fibrils and a lower proportion of amorphous protein matrix. This implies that it has a lower proportion of disulfide bonds compared to the paracortex. It is more porous and, therefore, more accessible to chemical treatments. In consequence, the ortho-cortex is affected by the different aggressions more quickly. This causes a loss of hair structure and leads to hair with a dull and lifeless appearance.

This method shows that **KERAMARE™** restores the integrity of hair fibers, repairing the damage caused by harmful factors, such as thermal treatments.

Using the same technique, the temperature of **water evaporation** is evaluated. The following chart (3) shows the temperature at which this evaporation takes place in the hair.

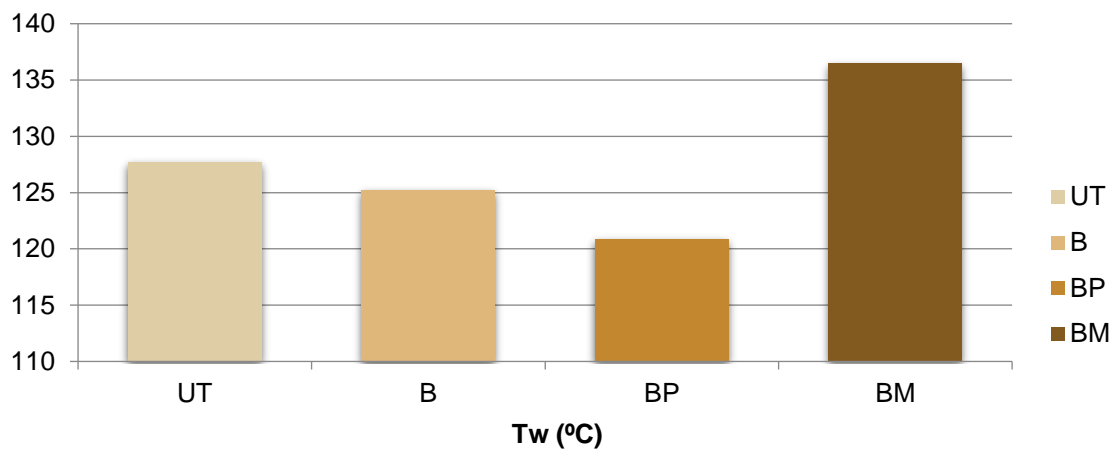


Chart 3. Water evaporation temperature variation,.

Hair contains in its structure a certain amount of retained water, which is a source of natural moisturization. At a certain temperature, water separates from keratin when it evaporates and hair is damaged. This can be caused by thermal treatments, such as the use of hair dryers and hair straighteners.

It can be seen that hair treated with **KERAMARE™** withstands higher temperatures:

- ✓ 8.8°C more compared to untreated hair
- ✓ 15.6°C more compared to placebo

With **KERAMARE™**, hair is protected and its moisturization is preserved against high temperatures or for a longer time. This is a benefit during the treatment with hot tools or other thermal or stressing processes to which hair is subjected.



**The application of KERAMARE™ substantially improves the integrity of the ortho-cortex, restructuring hair fibers and making them more resistant after the treatments that may damage hair**

## CONCLUSION

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**KERAMARE™** is an active ingredient developed to provide light, vitality and strength to hair fibers. All damage is repaired, hair is reborn and it shows its best appearance.

The application of **KERAMARE™**:

- ✓ Restructures keratin fibers
- ✓ Improves the integrity of the ortho-cortex. As a result, hair will be more resistant to harmful factors
- ✓ Protects hair against thermal treatments
- ✓ Improves the sensory and physical characteristics of hair, increasing brightness significantly
- ✓ Restores the natural appearance of hair, as it repairs the harmful effects of chemical treatments

## COSMETIC APPLICATIONS

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- ✓ Intensive strengthening and repairing hair products.
- ✓ Hair products for daily use (shampoo, mask, conditioner, foam, spray and serum).
- ✓ Thermal protectors.
- ✓ Fixation and hair styling products (spray, gel, hair spray and hair cream).
- ✓ Hair repairers for use after chemical and/or thermal hair treatments.
- ✓ Specific *products to increase hair brightness (serums, shampoos, leave-on masks)*
- ✓ Dyes.

## RECOMMENDED DOSAGE

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The recommended dosage is between 0.5 and 5%.



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