# A NEW BIOPOLYMER FOR REFRESHMENT

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# Summary

An innovative biopolymer known as the Rhizobian gum has been developed in France, which shows some dramatic refreshing effect on the skin.

The origin of this innovative project takes its source in the natural environment, and in particular the natural environment of the roots of sunflowers and wheat, where a symbiotic bacterium has been discovered. It is a Rhizobium bacterium, which is hosted by the roots, and which is able to synthesize a specific polymer showing a dramatic water binding capacity. This polymer is in particular synthesized in period of drought, and its biological role is to concentrate the small amount water present in the soil in order to take it available for the root, which becomes then able to absorb it.

This vital mechanism allows the plant to survive despite a severe climatic environment.

This basic research has been conducted in collaboration whit the French National centre of scientific Research (CNRS), and has lead to the isolation of the Rhizobium bacteria.

Rhizobian gum is a branched biopolymer consisting in the repetition of a polysaccharide unit of 3 molecules of glucose, 3 molecules of galactose and 1 molecule of glucuronic acid, whit one pyruvate group an average 1.6 acetyl groups.

The fresh effect of Rhizobian gum is a strong sensorial impact that 100 % of the consumers are able to perceive, and which is judged very pleasant by most of them. In addition to this, a large majority of consumers are perceived, and which is judge very pleasant by most of them. In addition to this, a large majority of consumers also feel a very pleasant relaxing sensation. Smoothness and softness are also felt by most consumers and qualified positively by most of them.

These qualities guarantee a strong impact on today's consumers.

# Introduction

The discovery of rhizobian gum comes from agronomical research, and the study of microorganisms living in the soil in symbiosis with the plants. A specific microorganism identified as a Rhizobium bacterium lives in contact with the roots of sunflower. This bacterium is gram-negative, catalase-negative, and oxydase-positive. In time of draught, the polymer synthesized by the

bacterium forms a film around the root. It stocks water present in small amount in the soil. The polymer has also an important role maintaining the soil structure.

As rhizobian gum plays the role of a natural water exchanger, it can be expected have interesting properties for cosmetic applications.

#### Materials and methods

# Production of the polysaccharide

#### Inoculation

The inoculum is grown from the flask to a 450-liter fermentor.

#### Fermentation

The inoculum is introduced in a 10m3 fermentor, where the production of the polysaccharide is carried out for 36-48 hours. At the end of the fermentation, the viscosity reaches 2000 mPa.s.

### **Purification**

The fermentation broth undergoes a heat treatment in order to confer gelling properties. This treatment also improves the elimination of the bacteria by centrifugation. The clear juice of centrifugation is filtered to eliminate the last particles. The clarified product is then precipitated by addition of ethanol. The powder is washed, dried and conditioned.

During the fermentation, rhizobian gum is produced at a temperature of 30°C. pH and air flow are significant parameters for the good development of the fermentation. During 10 hours, the culture is in exponential growth. rhizobian gum is a secondary metabolite: its production begins at the stationary phase. Glucose (20-25g/l) is consumed into 36 to 48 hours. The yield of the conversion of glucose into rhizobian gum is about 50%.

Figure 1

Measuring the viscosity allows to determine quickly the concentration of rhizobian gum in the medium.

Figure 2

#### Cosmetic evaluation

# Principle of the experiment

Gel formulations with 2% Rhizobian gum, with or without cornflower water, are poured in Petri dishes. Then gel rectangles are cut and applied on the lower eyelids (dark rings) of volunteers. Application time is 10 minutes.

After 10 minutes, color variation is measured in vivo using a chromameter.

# Principle of colorimetry

The chromameter allow

to characterize very accurately the color of the skin using 3 parameters: L\* (Lightness), C\* (Chrom a or Hue Intensity) and H (Hue Angle).

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C\* and H are obtained by the formulas  $C^* = \sqrt{a^{*2} + b^{*2}}$  and H=arctan b\*/a\*, where a\* is the red-green axis, and b\* is the yellow-blue axis of the L\*a\*b\* space.

According to the literature [1], C\* and H are the best to characterize skin coloration, Lightness L\* being too sensitive to experimental conditions (lighting conditions, chromameter probe pressure on the skin, etc.).

Figure 3: L\*a\*b\* space (Hue angle and intensity)

#### **Formulations**

#### Table 1

**Fabrication Process** 

Prepare the gel, then leave in a Bain Marie at 70 °C under agitation until homogenization Pour at 70°C in square Petri dishes (15x15 cm)

#### Measures

Measures are made at T=0 and T=10 min.

## **Results and Discussion**

# Characteristics of the polysaccharide

In its native form, Rhizobian Gum shows the characteristics of a viscous polysaccharide, after the heat treatment, the polysaccharide acquires gel properties.

The melting point of Rhizobian Gum is close to 60°C, but it gets lower after the sterilization. It can be close to body temperature.

#### Figure 4

Depending on temperature and concentration, the gel can shift between a dissociated flexible chains and partially organised chains with junction zones. The last conformation has a higher

affinity towards water. Figure 5 Cosmetic evaluation of the polysaccharide All the results obtained are significant (Student test) Effect of Rhizobian gum - Variation of the hue intensity after application of Rhizobian gum patches Figure 6 The hue intensity of dark rings is lowered by application of the patches. Rhizobian gum has a dark ring reduction effect. - Variation of the hue angle after application of Rhizobian gum patches Figure 7 The hue angle increases after application of the patches, showing an evolution of the hue from red to yellow-green. Rhizobian gum has an anti redness effect. Synergy with a decongestant: cornflower water - Variation of the hue intensity after application of Rhizobian gum patches containing cornflower water Figure 8 The hue intensity drops after application of the patches. Rhizobian gum presents synergetic effect with a decongestant such as cornflower water. - Variation of the hue angle after application of Rhizobian gum patches containing cornflower water Figure 9

The hue angle increases after application of the patches, showing an evolution of the hue from red to yellow-green. Rhizobian gum presents synergetic effect with a cornflower water as regards anti-

redness effect.

# Sensorial perception of patches of Rhizobian gum

After application, volunteers are asked to comment their sensorial perception of Rhizobian gum patches. For each of the qualities "Freshness", "Relaxing effect", "Smoothness", "Softness", they can choose a comment among "Extremely pleasant", "Very pleasant", "Slightly pleasant", "Neither pleasant or unpleasant", "Slightly unpleasant", "Very unpleasant".

Figure 10

## **Conclusions**

Rhizobian Gum improves the sensory properties of cosmetic formulations: it improves the brightness of emulsions and leaves a moisturizing, silky film on the skin.

Thanks to the particular properties of its gel, Rhizobian Gum has an outstanding self-cooling property. It is caused by the evaporation of water released during application.

This can be used to formulate mild cooling formulations, such as eye contour serums.

The efficacy of Rhizobian Gum patches against dark circle has been proved.

# References

[1] Ratan K. Chaudhuri and Cristina Hwang - Cosmetics & Toiletries magazine - Vol. 116, N°.9 (2001)

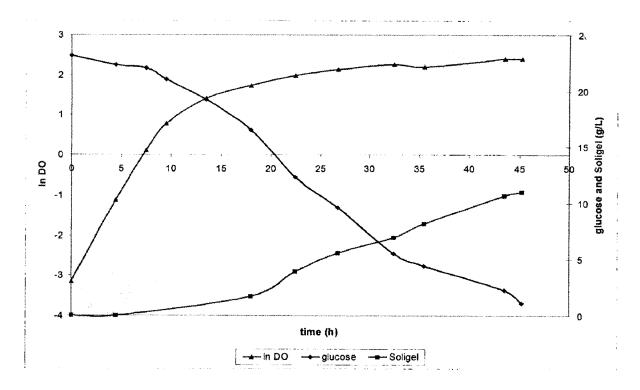
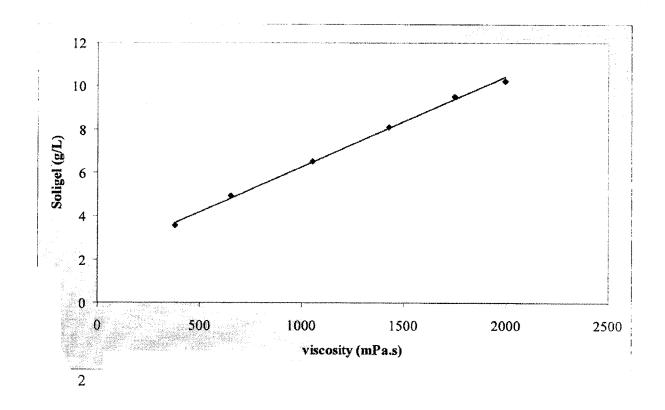


Figure 1



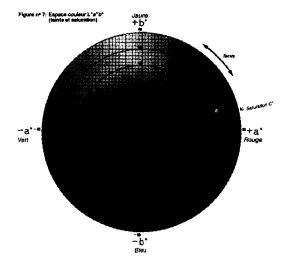
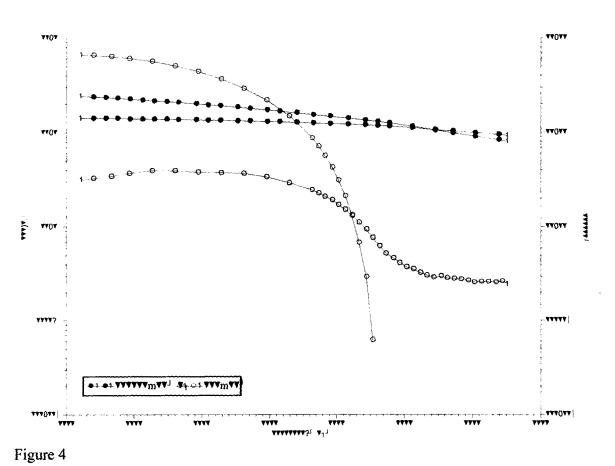


Figure 3



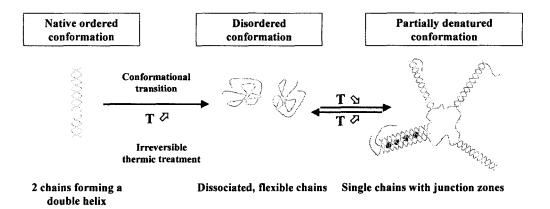


Figure 5

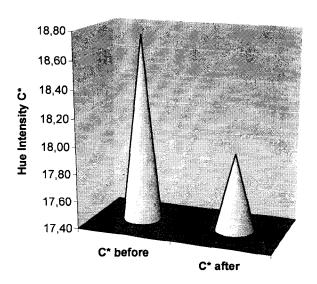


Figure 6

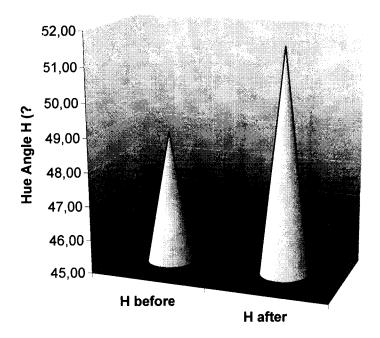


Figure 7

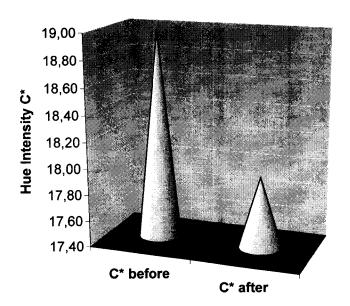


Figure 8

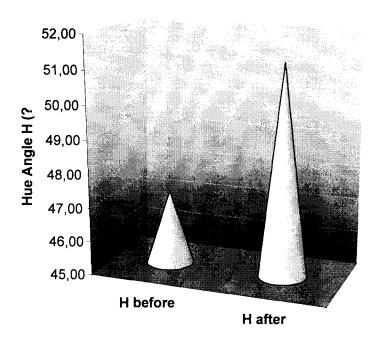


Figure 9

# Sensorial perception of SOLIGEL $\!\!^{\otimes}$ patches at 1 and 2% (30 people)

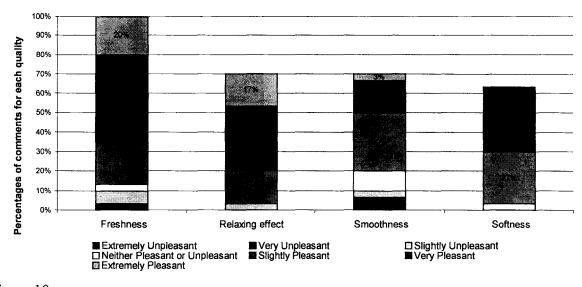


Figure 10

Phase gel	S 01-1	SB 01-2	
Rhizobian Gum	2%	2%	,
Osmosis-purified water	96.8%	/	
Cornflower Water	1	96.8%	
NaCl	1.2%	1.2%	
	100%	100%	

Table I