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N-Carboxymethyl chitosan in innovative cosmeceutical products

Riccardo Muzzarelli, Mirna Cucchiara* and Corrado Muzzarelli
Institute of Biochemistry, Faculty of Medicine, University of Ancona
*Sinerga srl, Italy

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Summary

A line of cosmetic products of interest for dental and oral applications has been prepared with N-carboxymethyl chitosan, endowed with biochemical and microbiological significance. Moreover, a systematic survey of the use of N-carboxymethyl chitosan in emulsions has been made, and an after-sun body milk is preseted as an example of advanced cosmetic formulations.

Riassunto

È stata preparata una linea di prodotti cosmetici funzionali di interesse per applicazioni odontostomatologiche e di igiene orale; le loro formulazioni comprendono N-carbossimetil chitosano, un polisaccaride avente capacità antimicrobica e riparativa. Inoltre, è stato fatto uno studio sistematico per l'uso dell'N-carbossimetil chitosano nelle emulsioni, e viene presentato un latte dopo-sole per il corpo, quale esempio di formulazioni cosmetiche avanzate.

INTRODUCTION

Chitin supports countless forms of terrestrial and marine life since many million years (Jollès and Muzzarelli, 1999). It is the most abundant nitrogen compound and cationic biopolymer. Thanks to modern technology, its isolation and purification from marine biomasses provides today chitosan, a natural biocompatible and biodegradable polymer approved for human consumption in Italy as well as other countries (Subasinge, 1999; Muzzarelli, 2000).

N-Carboxymethyl chitosan, originally developed by Muzzarelli (1982) (see also Muzzarelli et al., 1982) is the carboxymethylated soluhle form of chitosan, carrying a glycine aminoacid on most of its units. It belongs to a family of carboxymethylated chitins and chitosans, amply tested in the cosmetic and biomedical fields (Muzzarelli, 1988; Muzzarelli et al., 1989, 1994 a, b, 1998; Mattioli et al., 1999). Among its characteristic properties, the inhibition of the bacterial growth within the oral cavity, the prevention of Streptococcus mutans adhesion to the tooth surface, and the stabilization of the local pH, permit to recommend this functional ingredient for oral care products (Tarsi et al. 1998).

The antibacterial action of N-carboxymethyl chitosan combined with its efficacy against Candida albicans, qualify it as a functional ingredient in tootbpastes and mouthwashes.

N-Carboxymethyl chitosan keeps the filmogenic ability of the parent chitosan. The flow behavior and the viscoelastic properties of N-carboxymethyl chitosan aqueous system in the sol and gel domain have been investigated by means of dynamic, steady and transient shear techniques. These gel-like properties were explained in terms of the association of ordered chains to develop a cohesive network, analogous to that in normal gels but with weaker interactions between associating chains (i.e., weak gel) (Muzzarelli and Muzzarelli, 1998).

N-Carboxymethyl chitosan as a 1.0 % solution at pH 4.80 exerts a beneficial effect in terms of emulsion stability. It is a valuable functional ingredient of cosmetic hydrating creams in view of its durable moisturizing effect on the skin. The film-forming ability of N-carboxymethy chitosan assists in imparting a pleasant feeling of smoothness to the skin and in protecting it from adverse environmental conditions and consequences of the use of detergents (Muzzarelli and Muzzarelli, 1986; Chen et al., 2002).

N-Carhoxymethyl chitosan was found to be superior to hyaluronic acid as far as lasting hydrating effects for human skin are concerned (Muzzarelli et al., 1986).

EXPERIMENTAL

N-Carboxymethyl chitosan was prepared from shrimp chitosan as previously described by reductive amination of glyoxylic acid (Muzzarelli, 1997). The characteristics of the compound were checked by infrared spectrometry and H-NMR spectrometry; these techniques provided spectra from which the degree of carboxymethylation and the degree of acetylation were calculated. By 13C-NMR, the degree of acetylation is revealed by the hand at 24 ppm, while the degree of carboxymethylation and dicarboxymethylation are obtained with the analysis of the 53, 58 and 64 ppm bands. For the quality control purposes, the degree of acetylation is determined by i.r. spectrometry on the basis of the 1650 cm^{-1} band in the chitosan before the derivatization, while the degree of carboxymethylation is determined with the 1730 cm^{-1} band, typical of the carboxyl group.

The 1.0 % N-carboxymethyl chitosan solution including 1 % Germaben II is commercially available under the trade name Chitoglycan®. For certain preparations, freeze-dried N-carboxymethyl chitosan was prepared with a Heto Drywinner freeze-drier.

RESULTS AND DISCUSSION

Dental care

Four innovative formulations including N-carboxymethyl chitosan are presented here for the first time: a toothpaste deprived of abrasive inorganic compounds, a mouth wash, a gingival gel and an artificial saliva preparation that is often sought by dentists as well as for other clinical applications.

Tooth paste in gel form with no inorganics

For this preparation, in the turbo emulsifier the water is heated at 70°C and sodium saccharin, hydroxyethylcellulose and xanthan gum are added and dispersed in 15 min (Table 1).

Table I
Composition of a tooth paste with no inorganics.

N°	INCI NAME	%
1.	AQUA	q.b. 100
2.	SODIUM SACCHARIN	0.20
3.	HYDROXYETHYLCELLULOSE	1.50
4.	XANTHAN GUM	0.50
5.	SORBITOL	30.00
6.*	CITRUS GRANDIS, GLYCERIN	1.00
7.*	N-CARBOXYMETHYL CHITOSAN	5.00
8.	POLYSORBATE-20	0.80
9.	AROMA	0.20
10.	SODIUM LAUROYL SARCOSINATE	3.00

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The temperature is lowered to 30°C, and sorbitol, citrus grandis glycerin, N-carboxymethyl chitosan, sodium lauroyl sarcosinate are added; the homomixer is operated for 10 min.
Pre-mixed polysorbate-20 and aroma are added, to obtain a clear and homogeneous product. The pH may be adjusted with the aid of NaOH.

The final characteristics of the gel are the following: pH 8.22, viscosity at 25°C 6000 mPa.s with VT-02, pale yellow color, smell and taste typical of the aroma used.
In this gel sodium lauroyl sarcosinate is used as a foaming and antimicrobial detergent; its anionic nature is attenuated by its secondary amide function, and it is therefore compatible with Chitoglycan®. Xanthan and hydroxyethyl cellulose are two other polysaccharides used as rheological additives that impart viscosity and stability to the gel.
The rationale behind the use of Chitoglycan is to take advantage of its regenerative capacity in order to reduce inflammation and bleeding of oral tissues.

MOUTH WASH

This mouth wash is prepared by dissolving sodium saccharin in water in the turbo emulsifier; then citrus grandis glycerin and N-carboxymethyl chitosan are individually added and the emulsifier is operated to reach omogeneity (Table 2).

Table 2
Composition of a mouth wash.

N°	INCI NAME	%
1.	AQUA	q.b. 100
2.	SODIUM SACCHARIN	0.10
3.*	CITRUS GRANDIS, GLYCERIN	0.60
4.*	N-CARBOXYMETHYL CHITOSAN	5.00
5.	5-PPG-26-BUTETH26, PEG-40 HYDROGENATED CASTOR OIL	1.00
6.	AROMA	0.10

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Pre-mixed 5-PPG-26-buteth26, PEG-40 hydrogenated castor oil and aroma are then added and the homomixer is operated to reach homogeneity. The pH value may be adjusted with NaOH.

The final product is a clear and colourless solution with pH 5.20.
The rationale behind the use of Chitoglycan is to take advantage of its regenerative capacity in order to reduce inflammation and bleeding of oral tissues.

ARTIFICIAL SALIVA

The preparation includes the dispersion of hydroxyethyl cellulose in water at 70°C with the aid of the homomixer for 15 min (Table 3).

Table 3
Composition of artificial saliva.

N°	INCI NAME	%
1.	AQUA	q.b.100
2.	HYDROXYETHYL CELLULOSE	1.000
3.	SODIUM CHLORIDE	0.042
4.	POTASSIUM CHLORIDE	0.060
5.	MgCO3.6H2O	0.026
6.	CaCl2.2H2O	0.007
7.	K2HPO4	0.017
8.*	N-CARBOXYMETHYL CHITOSAN, SORBITOL	3.000

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Then, the inorganic salts are dissolved.
Finally freeze-dried NCMC is dissolved at 40°C.
The characteristics of the gel are: clear gel of pale yellow colour, pH 5.34, viscosity at 25°C 950 mPa.s with VT-02.
The gel is made of hydroxyethylcellulose and N-carboxymethyl chitosan. Sorbitol, already present as a plasticizer in the freeze-dried form of N-carboxymethyl chitosan, exerts a moistening action.
The salts are necessary to match the physicochemical and organoleptic characteristics of human saliva.

This artificial saliva is preservative free, to be packed in sterile environment and to be used in monodose applications.
The rationale behind the use of N-carboxymethyl chitosan is to take advantage of its mucoadhesive properties that ensure a long-lasting hydrating effect in the oral cavity.

GINGIVAL GEL

This gel is prepared with the aid of a homomixer by dispersing hydroxyethyl cellulose in water at 65°C for 15 min; then cooling at 40°C and adding the reduced sugars xylitol and maltitol to obtain a clear solution (Table 4).

Table 4
Composition of gingival gel.

N°	INCI NAME	%
1.	AQUA	q.b.100
2.	XYLITOL	10.00
3.	MALTITOL	10.00
4.	HYDROXYETHYLCELLULOSE	2.00
5.*	N-CARBOXYMETHYL CHITOSAN	2.00
6.	POLYSORBATE-20	0.35
7.	AROMA	0.15
8.	CHLOREXIDINE DIGLUCONATE	0.50

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Pre-mixed polysorbate-20 and aroma are then added.

Finally, the sorbitol plasticized freeze-dried form of N-carboxymethyl chitosan preliminarily dissolved in a minimum quantity of water is added, followed by chlorexidine digluconate.
The gel characteristics are: clear, pale-yellow gel, pH 5.00, viscosity at 25°C 15000 mPa.s with VT-02.
N-carboxymethyl chitosan is here used with the intention of providing relief to oral tissues who-

se functionality has been compromised by infections, or that have undergone surgical operations. Bleeding, inflammation and sour are promptly limited by N-carboxymethyl chitosan, particularly in the gingival pockets. This gel may find ample use at the end of periodical oral hygiene interventions.

EMULSIONS

After-sun body milk

Melt ceteryl alcohol, ceteryl glucoside, glyceryl stearate PEG-100 stearate, octyldodecanol, caprylic/capric triglyceride, ethylhexyl ethylhexanoate, dimethicone at 70°C, then add stearyl glycyrrethinate and bisabolol (Table 5).

In the homomixer warm water at 70°C and disperse guarhydroxypropyltrimonium chloride, add disodium EDTA, aloe barbadensis, glycerin and keep turbo on for 5 min.

Add the lipid phase to the water phase with turbo on for 10 min.

Cool to 50°C, add cyclopentasiloxane, dimethicone/vinyl dimethicone crosspolymer and use the turbo for 5 min and add perfume (~60%).

Cool further to 40°C, add perfume (~40%), and also propylen glycol, sophora japonica, N-carboxymethyl chitosan, hydrolyzed vegetable protein, menthyl lactate (ingredient menthyl lactate should be previously melted).

The characteristics of this white fluid emulsion are pH 4.55, viscosity at 25°C 5000 mPa.s with VT-02.

This emulsion includes a quaternary guar, various plant extracts, and therefore it should help reduce the unfavorable effects of sunshine on the skin thanks to the presence of Aloe barbadensis that protects against UV radiation and together with bisabolol exerts lenitive action.

The purpose of N-carboxymethyl chitosan in this formulation is to exert protection thanks to its filmogenicity against salt and adverse environmental substances, and help re-hydrate the skin.

Table 5
Composition of after-sun body milk.

N°	INCI NAME	%
1.	CETERYL ALCOHOL, CETEARYL GLUCOSIDE	2.50
2.	GLYCERYL STEARATE PEG-100 STEARATE	2.50
3.	OCTYL DODECANOL	5.00
4.	CAPRYLIC/CAPRIC TRIGLYCERIDE	7.50
5.	ETHYLHEXYL ETHYLHEXANOATE	7.50
6.*	DIMETHICONE	0.50
7.	STEARYL GLYCYRRETHINATE	0.10
8.	BISABOLOL	0.25
9.	AQUA	q.b.100
10.	GUAR HYDROXYPROPYLTRIMONIUM CHLORIDE	0.35
11.	DISODIUM EDTA	0.10
12.	ALOE BARBADENSIS	0.20
13.	GLYCERIN	2.00
14.*	PARFUM	1.00
15.*	CYCLOPENTASILOXANE, DIMETHICONE/VINYL	
	DIMETHICONE CROSSPOLYMER	2.50
16.*	PROPYLEN GLYCOL, SOPHORA JAPONICA	5.00
17.*	N-CARBOXYMETHYL CHITOSAN	10.00
18.*	HYDROLYZED VEGETABLE PROTEIN	10.00
19.	MENTHYL LACTATE	0.20

** Available from Sinerga*

CONCLUSIONS

N-Carboxymethyl chitosan is a versatile product that lends itself to a number of formulations. Besides the amply accepted emulsions already present in niche markets since 15 years, some innovative cosmetic formulations have now been prepared in order to take advantage from this functional ingredient. As a further extension of the emulsion range, an after-sun body milk has been presented, but, more important, a line of products suitable for dental/oral applications has been formulated.

The functional characteristics of N-carboxymethyl chitosan include hemostasis, ordered re-

construction of wounded tissues, anti-inflammatory and lenitive action, bactericidal/bacteriostatic action, candidacidal action, buffering capacity at physiological pH values, full biocompatibility, mucoadhesivity and biodegradability, that are definitely superior to those of other polysaccharides.

Being non-toxic, N-carboxymethyl chitosan can be used at any desired percentage in cosmetic and pharmaceutical applications, including percentages higher than those reported here.

Formulations of toothpastes have been developed including traditional alumina- or silica-based toothpastes, as well as advanced toothpastes deprived of inorganic ingredients.

N-Carboxymethyl chitosan is also recommended for mouthwashes, and especially for the treatment of pyorrhea or other gum diseases, where it improves effectively the health of gums and oral mucosae. Formulations undergoing pre-clinical studies include artificial saliva, and freeze-dried materials for the regeneration of gingival tissues.

A series of pre-systematic tests has been carried out with several representative formulations, in order to exploit the capacity of N-carboxymethyl chitosan to stabilize the emulsions. Final formulations of pre- and after-sun care products have been created and their organoleptic and stability characteristics have been found satisfactory.

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Author Address:

Prof. Riccardo Muzzarelli
Institute of Biochemistry
Faculty of Medicine, University of Ancona
Via Ranieri, 67
IT-60100 Ancona - Italy
Fax: +39 071 2204683
e-mail: muzzarelli@unian.it