Study of cosmetic applications with unmodified cellulose nanofiber as novel gel type ingredient

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ABSTRACT

Cellulose has been getting well known, since this material can be the best alternative of microplastics in all industrial field, especially in cosmetics field because of its biodegradability with OECD 301F and ASTM 6691D. Conventionally, in order to obtain cellulose nanofibers (CNF) having diameters of single-nano size, we must modify cellulose molecules with carboxylic and/or phosphoric groups. However, we have developed the novel process to obtain unmodified single-nano sized CNF (RCNF). The RCNF shows excellent transparency in the dispersion in water and the water dispersion shows relatively high under 1 wt% concentration and has high thixotropic properties for unmodified CNF. In addition, the RCNF film which obtained from the water dispersion after drying is very brittle, but by adding of butylene glycol (BG) in this water dispersion, the dried film ends up jelly type. This gel film can suppress water evaporation and chemicals penetration compared with RCNF film without BG. Moreover, we have made the evaluation of the emulsions with RCNF dispersion, the emulsions were stable without surfactants. At last, to address a possibility of RCNF in cosmetics, we prepared gel type formulation, and the formulations had excellent properties due to the high dispersibility and thixotropic properties of RCNF compared with the case of the conventional thickeners. In conclusion, the novel RCNF will be brilliant biodegradable ingredient for cosmetics.

KEYWORDS: cellulose nano fibers, natural ingredients, non-microplastic, film former, gel type formulation

INTRODUCTION

Cellulose nanofiber (CNF) is a plant-derived next generation material. It is nano-sized fiber produced through chemical and mechanical processes from wood, etc. It has a high specific surface area and such characteristics, high strength, high modulus of elasticity despite its light weight. In addition, this material shows significant biological properties, such as biodegradability, biocompatibility, and non-toxicity, namely being environmental friendly. Recently, microplastic contamination appears as one of the world's environmental main concerns[1], so CNF is getting more and more attention as a raw material which is not categorized as a microplastic.

There are some types of nano cellulose, (1) cellulose nanofiber, (2) cellulose nano crystal and (3) bacteria cellulose nanofiber. CNF is classified in micro fibrillated cellulose or cellulose nanofibrils[2]. CNF is generally defined as nano cellulose which has a width of 3-100 nm, length of 1 μm or more and a high aspect ratio and which is produced through a mechanical defibrating process[3]. It is difficult to make defibrillation of raw material cellulose. In order to do so, great deal of energy will be needed to obtain the appropriate size. Or by introducing either carboxylate or phosphate groups into cellulose, which is related to the electrostatic repulsion and/or osmic effects caused by those introduced functional groups[2], the defibrillation is carried out with gentle mechanical fibrillation. In cosmetic field, the modified CNF obtained by oxidation method is major, since the conventional unmodified CNF is clouded.

However, we have developed the novel process to obtain unmodified single-nano sized CNF (RCNF) and the RCNF shows excellent transparency in dispersing in water. The water dispersion using RCNF shows higher viscosity and better thixotropic property than that using conventional unmodified CNF. In addition, we found that the RCNF could be dispersed in mixture of water and BG with high concentration (in case of the water dispersion of commercialized modified CNF, the maximum concentration is around 2%).

The purpose of this study is to clarify these physical characteristics, and the various properties when applied in the cosmetics. At first, we analyzed a fiber diameter, pyrolysis temperature, dispersibility in the water and emulsion of RCNF and compared with CNF which prepared by other methods and other thickener. Second, we expected that RCNF would work as a barrier since it can form film on the skin, so we examined the potentials of RCNF film after drying. In addition, to address a possibility of RCNF in cosmetics, we prepared aqueous formulations.

MATERIALS and METHODS

MATERIALS

RCNF was provided by RENGO Co., Ltd. The following polyols were used; glycerin (Kao Corporation), butylene glycol (BG) (KH Neochem Co., Ltd.), pentylene glycol (Symrise Co., Ltd.). The followings are used for rheological, emulsifying and formulation experiment; xanthan gum (CP Kelco Inc.), arylates/C10-30 alkyl acrylate crosspolymer (Carbopol®, Lubrizol Corporation), sodium chloride (NaCl, FUJIFILM Wako pure chemical corporation), squalane (SOPHIM), silica treated ultrafine titanium dioxide (TiO_2) (DAITO KASEI Co., Ltd.), microcrystalline cellulose and cellulose gum (DAITO KASEI Co., Ltd.), hydroxypropyl methylcellulose stearoxy ether (DAIDO KASEI Co., Ltd.), carbon black water dispersion (DAITO KASEI KOGYO Co., Ltd.) and emulsion of styrene/acrylates copolymer (DAITO KASEI KOGYO Co., Ltd.). Seven Stars (tar: 14 mg, nicotine 1.2 mg, Japan Tobacco) was used a source of tobacco smoke as representative air pollution. Trifluoroacetic acid and NBD-Hydrazine (4-hydrazino-7-nitrobenzofurazanHydrazine), 4-(4,6-dimethoxy-1,3,5-triazin-2-yl)-4-methylmorpholinium chloride were purchased from Tokyo Chemical Industry Co. Ltd.

METHODS

Preparation of RCNF

Anion modified cellulose, which is prepared in the process of manufacturing cellophane from pulp, was defibrillated to form nanofiber while maintaining the Type I crystal structure, and then regenerated (converted) with heat or acid treatment to obtain non-substituted cellulose nanofiber (RCNF).

Size measurement of RCNF

After adjusting the concentration of RCNF dispersion, the specimens were stained and dried on a support film. Observations were made using a transmission electron microscope (TEM), and the fiber diameter of RCNF was measured from the images.

Pyrolysis temperature

The pyrolysis temperature was measured by increasing the temperature of freeze-dried RCNF from room temperature to 400 °C at a rate of 10 °C/min using a TG-DTA (TG8120, Rigaku Corporation). The temperature at the intersection of the tangent line at the time of large weight loss and the tangent line before weight loss was defined as the pyrolysis temperature.

Rheological properties

We analyzed the rheological behaviors depending on shear rate for each concentration and pH of RCNF dispersions using viscometers (TVB-10 or DVH-E, Tokimec Inc.). The concentrations of components in water were following ranges; from 0.3 to 1.0 % for RCNF, xanthan gum or Carbopol and from 0 to 5 % for NaCl. The pH of aqueous solution was adjusted in the range of pH 3 to pH11 with NaOH aqueous solution and HCl aqueous solution.

Emulsification properties

The emulsification potential of RCNF aqueous solutions was examined in different homogenizing conditions at room temperature. The concentration of squalane was 20%. 0.24% RCNF, xanthan gum or Carbopol aqueous solution was prepared by homogenizing with a homogenizer (MARK II 2.5, PRIMIX corporation) at 8000 rpm for 10 minutes. Squalane was mixed with the homogenizer at 8000 rpm for 5minutes. Emulsification features were evaluated as following parameters; the appearance and the stability.

Forming gel sheets

Several concentrations of RCNF and BG complex aqueous solutions were dried at 50˚C. We modified concentrations 0.5% for RCNF and from 0 to 10% for polyols in water. The gel sheets were immersed into the water to evaluate several properties. The mechanical strengths after immersion into water of these gel sheets were measured. For trapping effect experiment, RCNF gel sheet were prepared as follows: A piece of membrane filter (4.91 cm^2, 10 μm JH, Merck Millipore) was soaked in 1 ml of 0.5% RCNF aqueous solution and was then dried at 50˚C.

-Water evaporation property of gel film-

Water evaporation was measured with the apparatus modified hygrometer. The gel film was sandwiched between the plastic plates with hole. This sandwiched plate was set tightly on the bottle which was filled up water. The water in the bottle was wormed at 37˚C and kept at this temperature with the circulation of warmed water. The evaporation rate was measured as the change of humidity in hygrometer through the sandwiched plate for 60 minutes with 1 minute interval.

-Trapping effects of RCNF gel film against tobacco smoke-

The trapping effects of RCNF film against tobacco smoke were examined by determining aldehyde compounds (ACs) and benzo(a)pyrene (BaP) in de-ionized water diffused with tobacco smoke using fluorescence measurements. Tobacco smoke was diffused into de-ionized water after passing through membrane filters (4.91 cm^2, 10 μm JH, Merck Millipore) with or without RCNF gel film. De-ionized water (DIW) was reacted with 25 μM NBD-hydrazine in the presence of 0.05% trifluoroacetic acid for 30 minutes in the dark to determine the amounts of ACs. ACs were quantified by measuring fluorescence intensity (Ex; 480 nm, Em; 530 nm) using a microplate reader (Nivo, Perkin Elmer). BaP in the de-ionized water was quantified by measuring fluorescence intensity (Ex; 355 nm, Em; 460 nm).

Preparing formulation

Gel type sunscreen: Ingredients including each formulation in Table 3-A were homogenized with a homogenizer (MARK II 2.5, PRIMIX corporation) at 5000 rpm for 15 minutes. Add silica treated ultrafine TiO2 in Table 3-B into the premixture and mixed with the homogenizer at 5000 rpm for 10 minutes. We measured and compared sun protection factor (SPF) and protection grade of UVA (PA) of the prepared sunscreens using SPF-290AS automated UV transmittance/ SPF analyzer (Solar Light company).

Eyeliner: Each eyeliner in Table 4 was prepared in the same way as the aqueous sunscreen, replacing silica treated ultrafine TiO_2 with carbon black water dispersion and emulsion of styrene/acrylates copolymer. The bulk of the prepared eyeliner was filled into accordion-type eyeliner containers and compared writing quality such as color and amount of coming out when using the eyeliners. The accordion-type eyeliner container has been favored in recent years because of its good coloration and good bulk followability that can be used up to the end, although it is difficult to adjust the viscosity.

Viscosities of the prepared all samples were measured by a viscometer TVB-10M (Toki Sangyo Co., Ltd.) at room temperature.

RESULTS

Size measurement of RCNF

Fig. 1a shows a TEM image of RCNF aqueous dispersion, whose form of nanofiber consists with a diameter of 3 to 10 nm. Fig. 1b shows the appearance of aqueous dispersion of RCNF and mechanical fibrillated CNF (fiber diameter: approx. 10 to 100 nm) with 2 wt%, showing that RCNF is highly transparent.

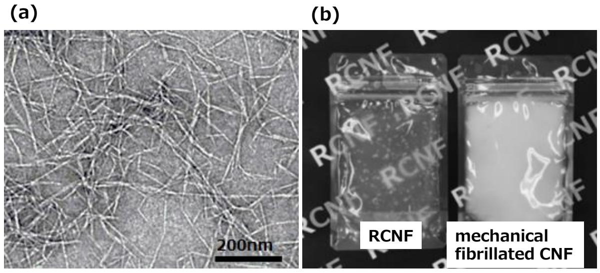

Fig. 1 Transmission electron micrograph (a) and appearance of water dispersion (b)

Pyrolysis temperature

The heat resistance of RCNF, and anion modified CNF as a comparison, is shown in Fig. 2. The anion modified CNF shows pyrolysis and gradual weight loss starting below 200 °C, while RCNF shows high pyrolysis stability, similar to that of pulp. The pyrolysis temperatures extrapolated from the TG curves are 256 °C for anion modified CNF and 313 °C for RCNF, indicating high thermal stability.

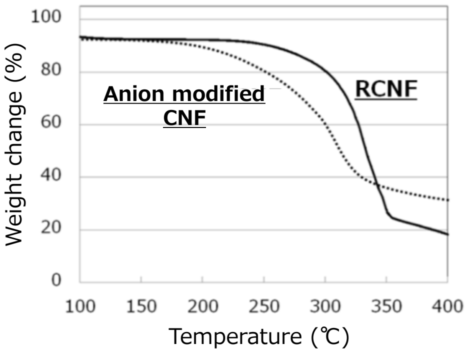

Fig. 2 Thermogravimetric analysis results

A graph was drawn plotting weight loss rate on the vertical axis and temperature on the horizontal axis, and the temperature at the intersection of the tangent line at the time of large weight loss and the tangent line before weight loss was defined as the pyrolysis temperature.

Rheological properties

Fig. 3a shows the relationship between viscosity and shear rate of aqueous dispersions of anion modified CNF and RCNF with 1 wt% at 20 ºC. Both anion modified CNF and RCNF show high viscosity in the low shear range. Getting higher shear rate the viscosity will be decreased dramatically, which means that these show thixotropic property. Anion-modified CNF, which is close to fully nano-dispersed, shows a strong thixotropic property than RCNF, but RCNF has high thixotropy for a mechanical fibrillated (non-modified) CNF. Fig. 3b shows the viscosity change with addition of NaCl (NaCl 0% = 100). Xanthan gum was almost completely unaffected by NaCl, whereas RCNF showed the decrease in viscosity. Carbomer had no NaCl tolerance at all. Fig. 3c shows the viscosity change with pH as a percentage of pH 7 (pH 7 = 100). RCNF showed the highest viscosity at pH 5 and decreased at pH 3 and pH 11. Xanthan gum showed a slight decrease in viscosity at pH 3 but had almost no effect. Carbomer did not thicken at all in acidic conditions.

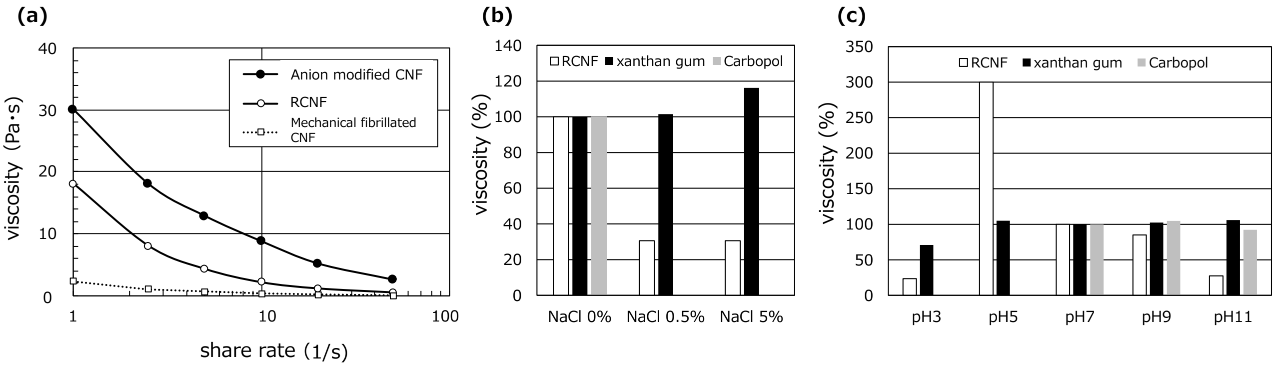

Fig. 3 Influence of each parameter on RCNF viscosity

(a) Relationship between shear rate and viscosity of anion-modified CNF, mechanical fibrillated CNF and RCNF as 1 wt% (b) Viscosity change when the viscosity of 0.3 wt% each thickener (RCNF, xanthan gum and Carbopol) without NaCl added is 100 % (c) Viscosity change when the viscosity of 0.3 wt% each thickener at pH 7 is 100 %

Emulsification properties

The emulsification properties in Table 1 were evaluated. The composition of RCNF/ de-ionized water/ squalane with a weight ratio of 0.24/ 79.76/ 20.00 is the most suitable to obtain the stable O/W emulsion without surfactants (Table 1). For Carbopol, we could make the same emulsion as the case of RCNF. However, we could not obtain the stable emulsion for Xanthan gum. The emulsions of RCNF are stable for the pH range 5-11. In comparison with the case of Carbopol, the stable pH range is 7-11. However, in addition of NaCl, the stability of the emulsion was getting less.

Table 1 Emulsification property

Change added	RCNF	Xanthan gum	Carbopol
(Emulsion type)	o/w	failed	o/w
Add 0.5% NaCl	failed	failed	failed
pH 3	failed	failed	failed
pH 5	emulsified	failed	failed
pH 7	emulsified	failed	emulsified
pH 9	emulsified	failed	emulsified
pH 11	emulsified	failed	emulsified

The influence of 0.5% NaCl addition and pH on emulsification of RCNF, xanthan gum or Carbopol at 0.24%, water at 79.76% and squalane at 20% was tested.

Gel Film properties

When 0.5 wt% of the water dispersion on RCNF is dried for 18 hours at 50℃, film is obtained. However, the film is very brittle (Fig. 4a). On the other hand, by adding of BG, pentylene glycol or glycerin in this water dispersion, the dried film ends up jelly type (Fig. 4b). Jelly property will increase with the concentration of polyols. Therefore, to verify the properties of the gel sheet prepared with several BG content, we examined the effect on water evaporation and trapping effect on chemicals including tobacco smoke as an environmental stimulus. BG was chosen as a polyol in the examination, since we considered that substantial benefits to the skin as the result of the gel film prepared with RCNF-BG aqueous solution could be estimated. Moreover, BG is widely used in cosmetic formulations compared with other polyols. Gel film made from RCNF-BG aqueous solution significantly suppressed water evaporation compared with the control (Table-2). To examine the trapping effects of RCNF gel

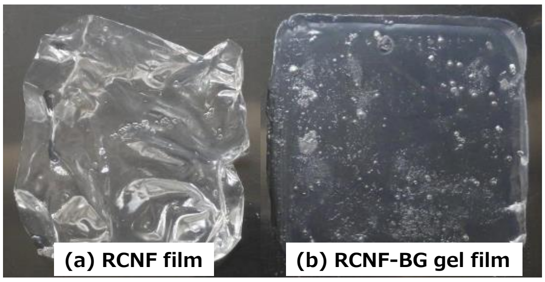

Fig. 4 Image of RCNF film(a) and RCNF-BG gel film(b)

Table 2 Water evaporation through gel-sheets at the initial state

	Water evaporation rate (%/minute)	significance vs control	significance vs only RCNF
Control	4.66±0.896	-	-
only RCNF	3.72±0.425	ns	-
RCNF+5%BG	2.98±1.329	ns	ns
RCNF+10%BG	2.42±0.407	*	*

A water evaporation rate was calculated as humidity changes per minute. Control showed water evaporation rate without RCNF sheet in the measurement condition. Measurements were carried out in triplicate, results were expressed mean ± standard deviation (S.D.). Student t-test. Significance: * $p<0.05$.

film, the amounts of ACs and BaP in DIW diffused with tobacco smoke passed through membrane filters treated with or without RCNF were quantified. RCNF-BG aqueous solution treated membrane filters showed excellent trapping effects for both ACs and BaP (Fig. 5). More than 90% of ACs and BaP were trapped in the RCNF-10% BG aqueous solution treated membrane filter compared with the control (non-treated membrane filter). In both examinations, the gel film from RCNF and BG showed significant superior effects to that from RCNF (Table 2, Fig. 5).

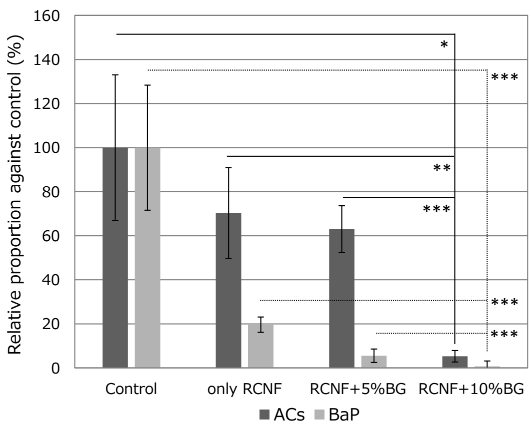

Fig. 5 Trapping effects of RCNF-treated filters on ACs and BaP in tobacco smoke

Each value represents the mean ± SD of three experiments. T-test, **p*<0.05, ***p*<0.01, ****p*<0.001. Trapping efficiency is expressed as a percentage versus the value of the non-treated membrane filter diffused with tobacco smoke (Control).

Possibility for new formulation

Gel type sunscreen: The sunscreen containing RCNF show very good SPF performance compared with the case of the conventional thickeners such as xanthan gum and Carbopol (Table 3). The formulation of RCNF has the lowest viscosity among other formulas, on the other hand the silica treated TiO_2 was well dispersed in that of RCNF and the RCNF could form a uniform film due to BG, so it showed highest SPF value.

Eyeliner: The bulk on eyeliner containing xanthan gum and commercial product blurred after 50 writings. However, the eyeliner containing RCNF did not blur and maintained a smooth writing quality (Table 4). The commercial product was contained also xanthan gum, BG, carbon black and styrene/acrylates copolymer, and it is similar in composition to the prepared xanthan gum formulation and the base of the formulation. When RCNF is applied in eyeliner, it is possible to prepare formulas that can be used up to the end without bulk blurring even after writing many times.

Table 3 Formulation of aqueous sunscreen

	Ingredients	RCNF	Other cellulose	Xanthan gum	Carbopol
A	De-ionized water	80.75	84.20	85.70	86.40
A	RCNF	1.00	-	-	-
A	microcrystalline cellulose and cellulose gum	-	2.00	-	-
A	hydroxypropyl methylcellulose stearoxy ether	-	0.15	-	-
A	xanthan gum	-	-	1.00	-
A	arylates/C10-30 alkyl acrylate crosspolymer	-	-	-	0.20
A	AMPD	-	-	-	0.10
A	BG	7.95	3.35	3.00	3.00
A	Preservative	0.30	0.30	0.30	0.30
B	silica treated ultrafine TiO_2	10.00	10.00	10.00	10.00
	Viscosity (Pa･s)	**49.2**	**92.2**	**50.7**	**72.3**
	SPF/PA （UVA-PF)	**26.99/ PA++++ (22.05)**	**22.13/ PA++++ (17.81)**	**9.85/ PA+++ (8.42)**	**17.38/ PA+++ (13.99)**

The preparing formulations and measurement of viscosity and SPF/PA were carried out with methods described in material and methods.

DISCUSSION

In the study, we found that RCNF, developed the novel process to obtain unmodified single-nano sized CNF, has excellent thinness, transparency, and rheological properties as a nanofiber made of pure cellulose without chemical modification, and it has pyrolysis stability similar to that of pulp (Fig. 1, 2 and 3). In addition, RCNF could emulsify oil stable without surfactant (Table 1). It is well known that anion modified CNF can emulsify oils without surfactant, due to combination of its network structure of CNF, the improvement of viscoelasticity of the emulsion interface, and the electric double repulsion caused by the introduced functional groups. However, the stable emulsification can be achieved even with RCNF without functional groups indicates that the RCNF is well dispersed to a level close to that of anion modified CNF, although the electrostatic repulsion is reduced, and some agglomeration occurs as a result of regeneration.

Table 4 Formulation of eyeliner

Ingredients		RCNF	Xanthan gum
A	De-ionized water	57.55	58.50
A	RCNF	0.20	-
A	Xanthan gum	-	0.20
A	BG	1.95	1.00
A	Preservative	0.30	0.30
B	Carbon black water dispersion	20.00	20.00
B	Emulsion of styrene/acrylates copolymer	20.00	20.00
	Viscosity (mPa·s)	**356**	**320**
Writing quality	RCNF / Xanthan gum / Commercial product 		

The preparing formulations and measurement of viscosity were carried out with methods described in material and methods. The writing quality of the eyeliners were judged with 50 writings on paper as shown in the image.

RCNF formed a gel-like film (gel-film) in the presence of polyols and the jelly properties, thickness and softness, were increased depending to concentration of polyols. Since CNF is known to make very light and strong film, the following effect were expected to RCNF gel film when formed on the skin: suppressing water evaporation from the inside and acting as a barrier against external stimuli from the outside. The gel film prepared with RCNF-BG aqueous solution was able to suppress water evaporation by half compared to the control, but there was no significant difference between gel-film prepared with 5 % BG and 10 % BG (Table 2). Tobacco smoke was used as an external stimulus, representative air pollutant that has become a problem in recent years, to examine the trapping effect of RCNF gel film. RCNF gel film containing 10 % BG showed excellent trapping effects both for ACs and for BaP (Fig. 4). Gathering results, it seems that BG, well known as a humectant, exist in the RCNF by holding water, and the film become jelly like. However, it is unlikely that RCNF interacts strongly with water, and the amount of water that can be held by BG is limited, so RCNF gel film could not be completely suppressed water evaporation. On the other hand, the trapping effect of RCNF gel film against external stimuli was very effective, since RCNF could form a uniform film due to presence of BG.

CNF gives such superior in durability enough to be used in a variety of applications as a composite material[6], and once it becomes a film after dry, it will not recover to a gel-like state again. However, we found that the RCNF could be re-dispersed in water easily depending on the ratio of BG, RCNF, and water, and successfully prepared condensed RCNF which 8 times the concentration of conventional modified CNF aqueous dispersions (up to 2%)[5]. By using condensed RCNF, it may be possible to achieve cosmetic formulations with a high concentration of CNF, which was not possible with the 2% CNF aqueous dispersion until now. Formulations containing 1% RCNF as shown in Table 3 can also be prepared without any restrictions on ingredients, and as evidence of its excellent dispersibility, it has a higher SPF than formulations using other thickeners. Furthermore, it is possible to prepare excellent formulations such as the eyeliner formulated with RCNF in Table 4, which maintains some viscosity, but its thixotropic properties make it writing quality.

CONCLUSION

As shown so far, RCNF is extremely thin and well dispersible as a pure cellulose nanofiber. A feature different from modified CNF, high thermal stability, is useful because it prevents alteration due to heat or long-term storage. In conclusion, the novel RCNF will be very lucrative ingredient for cosmetics, especially gel type cosmetics such as skin care and make-up, and as a biodegradable ingredient.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

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