Preparation of Low-Protein Natural Rubber Latex: Effect of Polyethylene Glycol

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ABSTRACT: Low-protein content natural rubber latex was produced by using a nonionic surfactant-polyethylene glycol (PEG). Extractable protein content of natural rubber latex was found to decrease with PEG treatment and reduction increased with increase in the molecular weight of PEG. The low-protein latex samples were characterized by tensile testing, Fourier transform infrared and thermogravimetric analysis. The results have shown 35% reduction in the extractable protein content, without any compromise on the mechanical properties of the latex; however, thermal stability of low-protein latex was found to be reduced marginally with PEG treatment. © 2009 Wiley Periodicals, Inc. J Appl Polym Sci 114: 806–810, 2009

Key words: aging; films; latices; mechanical properties; proteins

INTRODUCTION

The applicability of gloves and other latex products is restricted because of allergic problems associated with natural rubber latex.[1,2] It has been reported that some of the proteins present in the latex are mainly responsible for the allergic reactions.[3–11] Significant reduction in the allergic response of natural rubber latex can be achieved by the reduction in its protein content, however, out of the total proteins present in the latex or latex film only a fraction is extractable.[12]

Several techniques are available to reduce the protein content of the latex, such as leaching, autoclaving, chlorination, use of proteolytic enzymes, and use of nonionic surfactants. Leaching is effective only when we do that process for a few hours and so it is not commercially viable in reducing protein content to a greater level for production of gloves, catheters, etc. Steam autoclaving can affect physical properties unless precautionary measures are taken at the compounding stage. The use of chlorination may affect the strength of the gloves and it reduces the color of the gloves. Proteolytic enzymes are proteins and so we cannot rule out the possibility of them leading to a new allergy.[13] Moreover, a long incubation time is needed for enzymatic deproteinisation.[14] But the use of nonionic surfactant[15,16] is a comparatively better method and it will not affect the mechanical properties to a greater extent. This article describes a method for protein reduction by using a nonionic surface active material and subsequent characterization of low-protein rubber latex.

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EXPERIMENTAL

Materials used

Natural rubber latex of dry rubber content (DRC) 32.7% was procured from M/s Wynad resins, Kerala. Polyethylene glycol (PEG) was obtained from SD Fine Chemicals, Mumbai, India. Sodium chloride, potassium dihydrogen phosphate, hydrated disodium hydrogen phosphate, potassium chloride, sodium carbonate, sodium hydroxide, cupric sulfate pentahydrate, and sodium deoxycholate (DOC) were procured from E.Merck India, Mumbai. Follins reagent and trichloroacetic acid (TCA) were obtained from Merck Specialities, Mumbai, India. Sodium tartarate was obtained from Qualigens Fine chemicals and phosphotungstic acid (PTA) was obtained from Loba Chemie, Mumbai. Standard protein solution was albumin from chicken egg white grade V (A-5503) and it was procured from Sigma–Aldrich, USA.

Preparation of low-protein latex by treating with PEG

To the field latex was added 10% aqueous solution of PEG to get a concentration of 0.2% (w/w). It was

TABLE I
Formulation for Compounding Latex to Produce Gloves

Constituents	Concentration (phr)
Latex	100
Sulfur	1.1
Zinc oxide	0.35
ZMBT	0.2
ZDEC	0.85
TiO_2	0.25
Wingstay L	0.75
KOH	0.15

allowed to mature for 24 h and then centrifuged at an rpm of 7129 in a centrifuging factory. After that it was casted on to a glass tray to prepare the film. The film was leached in hot water kept at a temperature of about 80°C for about 2 min. A known weight of the sample was taken and protein test was done by modified lowry method as per ASTM D 5712 : 99. Different molecular weights of PEG were added to the latex and their effect on the reduction of protein was studied. After optimizing the molecular weight, concentrations of PEG were varied from 0.1% to 0.3% and their efficiency was studied by making gloves using the three latices.

Determination of protein content

A known weight of the test specimen was extracted with a dilute extraction buffer of ratio 1 : 9 (buffer : water). The pH of the buffer solution was adjusted to 7.4 ± 0.2. The test specimen was immersed in the extraction buffer solution. The quantity of the extraction solution was between 5 and 10 mL per 1 g of the glove material or the latex film. Extraction was done at 25 ± 5°C for 120 ± 5 min. The test specimen was removed from the extraction solution and the remaining solution was centrifuged to 15 min at 1865 rpm. Then 4 mL each of the reagent blank, standard protein solution, and the specimen extract were transferred into a polypropylene tube. A duplicate specimen extract was also taken. A total of 0.4 mL sodium deoxycholate (DOC) was added, mixed well, and kept for 10 min. A total of 0.8 mL of freshly prepared solution of 50 : 50 TCA and PTA was added and the protein was precipitated as acid precipitate. The contents were mixed well and allowed to stand for 30 min. The acid precipitate was centrifuged at 6236 rpm for 15 min.

A total of 1.2 mL of 0.2 N NaOH solution was added to each tube, including blank so as to redissolve the precipitated protein and shaken well so that the protein was completely redissolved to a clear solution. A total of 2.5 mL of reagent C (alkaline copper tartarate solution) and C′ (alkaline tartarate solution) were added, respectively, to the specimen extract and duplicate. The solution were mixed well and kept for 15 min at room temperature. A total of 0.3 mL of 50% follins reagent was added to each of them and thoroughly mixed immediately. Then both were kept for 30 min at room temperature.

TABLE II
Properties of Modified Lattices

Type of Latex	DRC (%)	MST (s)	NH_3 content (%)
Latex without PEG treatment	60.1	960	0.3
Latex treated with PEG 4000	60.05	960	0.25
Latex treated with PEG 6000	60.05	960	0.25
Latex treated with PEG 9000	60.05	970	0.25
Latex treated with PEG 20000	60.05	1010	0.21

The final assay mixture was transferred to a cuvette and the concentration of the standard solution C and C′ were measured.

$$\text{Extractable protein content, } E.P = \frac{C \times V \times F}{S}$$

where C is the protein concentration of extract in μg/mL, V the volume of extraction buffer in mL, F the dilution factor, and S the surface area in dm^2 of the NR specimen, i.e.

$$\frac{\text{length(mm)} \times \text{width(mm)} \times 4}{10,000}$$

Characterization

Characterization of the optimized samples was done by using Fourier transform infrared (FTIR) spectrometer (Bruker, Tensor 27) and Thermogravimetric Analyzer (Q50, TA Instruments).

Production of gloves using treated latex

Gloves were made using 0.1, 0.2, and 0.3% PEG (molecular weight 20,000) containing latices. The latices were compounded using the formulation given below in Table I. Gloves were produced by first dipping the preheated former in compounded latices containing PEG for 40 seconds. All other parameters required to make gloves were kept constant as factory conditions.

TABLE III
Protein Content of Uncompounded Lattices

Latices used	Protein content (microgram per gram)
Latex without Surfactant	49.14
Latex with PEG(mol. wt 4000)	47.29
Latex with PEG(mol. wt 6000)	38.89
Latex with PEG(mol. wt 9000)	35.02
Latex with PEG(mol. wt 20,000)	32.13

TABLE IV
Properties of Latex Without Surfactant Treatment and Latices Treated With 0.1, 0.2, and 0.3% PEG

Properties	Latex Without surfactant treatment	0.1%PEG	0.2% PEG	0.3%PEG
Total solid content (%)	61.4	64.6	64.65	64.66
Dry rubber content (%)	60.1	63.3	63.2	63.3
Ammonia content (%)	0.3	0.3	0.3	0.3
VFA number[a]	0.041	0.047	0.047	0.05
KOH number[b]	0.5	0.5	0.5	0.5
MST (s)	960	1000	1010	1030

[a] VFA number (volatile fatty acid number)—the number of grams of potassium hydroxide equivalent to the anions present as salts of steam-volatile acids in a quantity of latex, which contains 100 g of the total solids.
[b] KOH number—the number of grams of potassium hydroxide equivalent to the anions present as ammonium salts in a quantity of latex which contains 100 g of total solids.

Tensile properties

Tensile properties were performed on Shimadzu autograph AG1 series as per ASTM D412 at a crosshead speed of 500 mm/m.

RESULTS AND DISCUSSION

The properties of the modified latices like DRC, mechanical stability time (MST), and ammonia content are given in Table II. It is clear from the MST values that the latex is stabilized by the addition of PEG and it is higher for the latex treated with PEG having molecular weight 20,000. Table III shows the protein content of latex without surfactants and latices treated with different molecular weights of PEG. Proteins get adsorbed on PEG. It is clear from Table III that enhanced reduction in protein content is observed when PEG of higher molecular weight is treated with latex. This may be due to the increase in hydrogen bonding owing to the increase in oxygen atoms in the repeating units of PEG with higher molecular weights. The PEG–protein moiety can be removed from latex by centrifugation. Commercial viability cannot be achieved if we increase the molecular weight beyond 20,000 because of the high cost of PEG for its higher molecular weights. Therefore, because of the higher protein reducing capacity and mechanical stabilization, PEG having molecular weight 20,000 was taken for further study.

Table IV shows the properties of centrifuged latex without PEG treatment and latices treated with 0.1, 0.2, and 0.3% PEG having molecular weight 20,000. It can be seen that the latex properties are more or less uniform for latices with and without PEG. Table V shows the extractable protein content of gloves prepared (as per formulation given in Table I) from latex concentrated with 0.1, 0.2, and 0.3% PEG (molecular weight 20,000) and that of centrifuged latex without PEG. It is clear from the table that 0.2% PEG effectively reduces the protein content compared with glove produced from centrifuged latex without PEG treatment, whereas 0.3% PEG shows only a very marginal decrease. Moreover, the cost will be increased if we use 0.3% PEG.

Required chloroform number is between 3 and 4. (It is an arbitrary number that is assigned to the latex on the basis of appearance. Four stages of prevulcanisation are usually distinguished by this test and are assigned as follows:

Chloroform number 1—coagulum is a tacky mass, breaking in a stringy manner when stretched.
Chloroform number 2—the coagulum is a weak lump, which breaks short when stretched.
Chloroform number 3—the coagulum has the form of a nontacky agglomerate.
Chloroform number 4—the coagulum has the form of small dry crumbs.)

Latex with 0.2% PEG attained the chloroform number within 48 h. But it takes 96 h in the case of latex with 0.3% PEG. Therefore, for further studies, latex prepared with 0.2% PEG (molecular weight 20,000) is selected.

TABLE V
Protein Content of Gloves Prepared from Latices of Different Percentages of PEG (Molecular Weight 20,000) and from Latex Without Surfactant Treatment

Latex used	Proteins ($\mu g/dm^2$)
0.1% PEG	57.05
0.2% PEG	47.93
0.3% PEG	47.96
Centrifuged latex without PEG	85.01

TABLE VI
Tensile Properties of Gloves Before and After Aging Prepared by Treating Latex With 0.2%PEG

Before aging			After aging		
Stress at 500% elongation (MPa)	Elongation at break (%)	Tensile strength (MPa)	Stress at 500% elongation (MPa)	Elongation at break (%)	Tensile strength (MPa)
2.24	940	24.5	2.7	910	22.6
2.27	940	24.9	2.65	910	22.7
2.27	940	24.9	2.65	910	22.4

TABLE VII
Specifications of Mechanical Properties of Surgical and Examination Gloves

	Before aging			After aging	
Type of glove	Stress at 500% elongation (MPa)	Elongation at break (%)	Tensile strength (MPa)	Elongation at break (%)	Tensile strength (MPa)
Surgical	<5.5	≥750	≥24.0	≥560	≥18
Examination (type I)	2.8–5.5	≥650	≥18	≥500	≥14
Examination (type II)	<2.8	≥650	≥14	≥500	≥14

TABLE VIII
Tensile Properties of Commercial Gloves

Before aging			After aging	
Stress at 500% elongation (MPa)	Elongation at break (%)	Tensile strength (MPa)	Elongation at break (%)	Tensile strength (MPa)
2.34	923	25.52	812	22.0
2.97	925	25.74	864	21.44
3.1	920	26.01	849	22.2

Tensile strength of the gloves prepared by using 0.2% PEG before and after aging is found out by cutting dumbbell shaped samples from the gloves and it is found that it meets the specification requirements of both examination (type II) and surgical gloves. The tensile properties before aging and after aging (at 100°C for 22 h in a hot air oven) are given in Table VI. Specifications of surgical and examination gloves are given in Table VII as per ASTM D 3577 and ASTM D 3578, respectively. Tensile

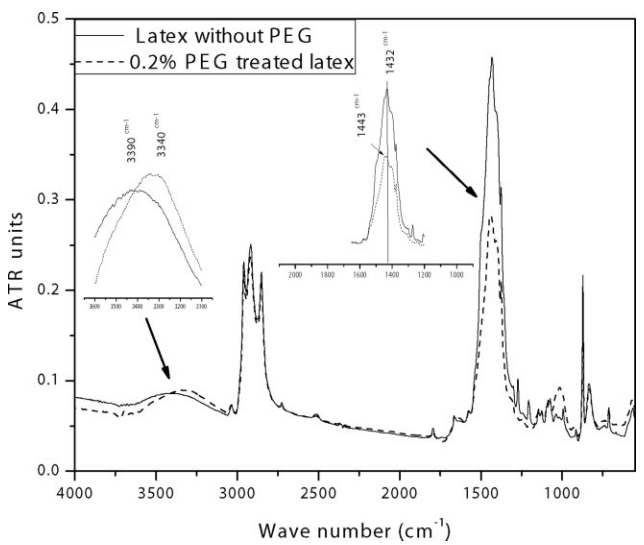

Figure 1 Infrared spectra of latex without PEG treatment and latex with 0.2% PEG treatment.

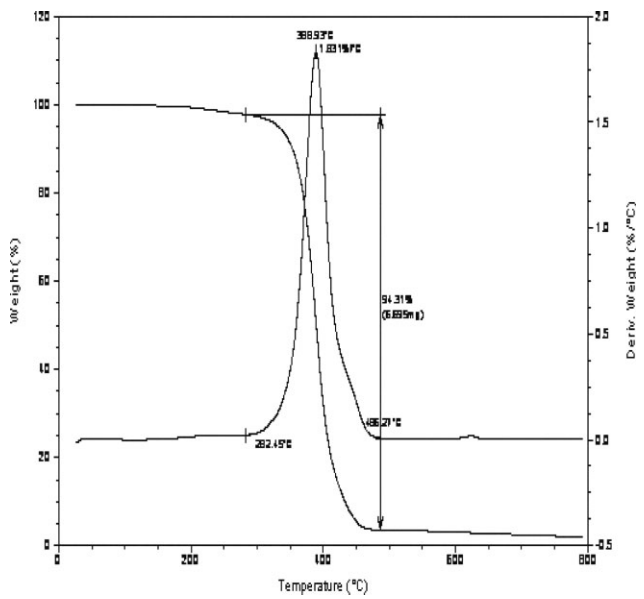

Figure 2 TGA curve of centrifuged latex without PEG treatment.

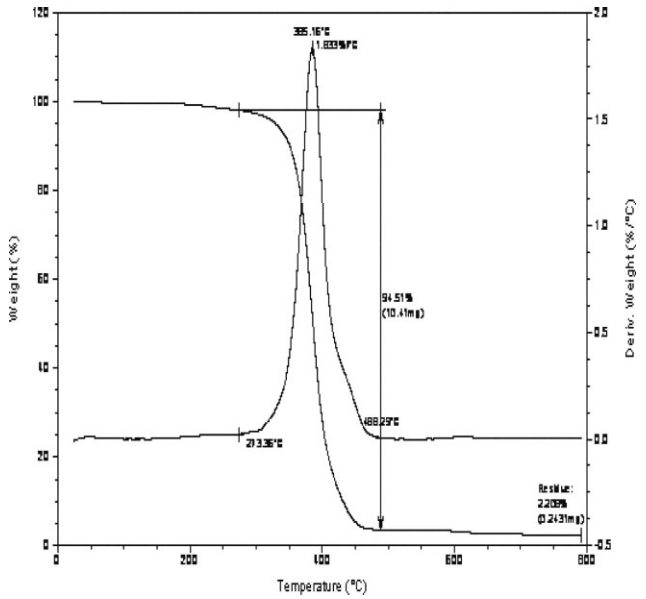

Figure 3 TGA curve of 0.2% PEG-treated latex.

properties of commercial gloves are given in Table VIII. FTIR spectra of the aforesaid low-protein latex (compounded) film and that of compounded latex film without PEG treatment were shown in Figure 1. For the compounded latex film without PEG, a peak at 3390 cm^{-1} is observed, which shifted to 3340 cm^{-1} in the case of 0.2% PEG-treated latex. The lower frequency shift is due to the reduction in peptide linkage, which originates from the protein present in the latex. The peak at 1432 cm^{-1} corresponding to the —NH deformation (in latex without PEG treatment) is shifted to 1443 cm^{-1} in the case of 0.2% PEG-treated latex and also the intensity of the peak is reduced considerably indicating the reduction in free —NH concentration, which, in turn, indicates the reduction in protein content. The peak present at 1015 cm^{-1} in PEG-treated latex shows the presence of ether linkage due to the presence of traces of PEG in the latex.

Figures 2 and 3 shows the TGA thermogram of centrifuged latex film without PEG and PEG-treated latex film respectively. In the case of latex without PEG treatment, the degradation starts at 282.4°C but it is shifted to 273.4°C in the case of PEG-treated latex. Peak degradation temperature of latex film without PEG treatment is 388.9°C with rate 1.83%/°C and that of PEG-treated latex film is 385.2°C with rate 1.83%/°C. Hence, the addition of PEG slightly reduces the thermal stability of the latex film.

CONCLUSIONS

Low-protein latex was successfully prepared with PEG. About 35% reduction in the extractable protein content of natural rubber latex was observed when PEG was incorporated. The reduction in extractable protein content was found to be increased with increase in PEG (molecular weight 20,000) up to 0.2%, but did not show significant increase on further increase in PEG concentration. The FTIR spectra also confirm significant reduction in the protein content after PEG treatment. The mechanical properties of the gloves produced from low-protein latex were found to be similar to that of commercial latex gloves.

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