Latex 2006 24-25
January 2006 – Frankfurt, Germany

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TECHNOLOGICAL AND PHYSICAL PROPERTIES OF A NEW, LOW ANTIGENIC PROTEIN NATURAL RUBBER LATEX

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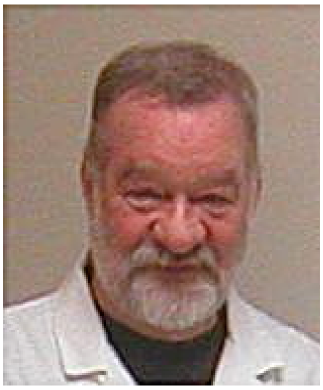
BIOGRAPHICAL NOTE



Travis W. Honeycutt is the founder and Chairman of Vystar Corporation. Mr. Honeycutt has a distinguished record of developing and commercializing new products by applying chemical engineering. Prior to Vystar, Mr. Honeycutt founded Isolyser Company, Inc., which developed and marketed products to the healthcare industry. Isolyser (currently Microtek Medical Holdings) grew from a start up to a publicly traded company with approximately $180 million in annual sales. Mr. Honeycutt retired as Executive Vice President in 1999 to pursue other ventures. Mr. Honeycutt holds more than one hundred patents and applies them to commercialize a wide range of technologies. Mr. Honeycutt has a Bachelor of Science from North Carolina State University, a Masters Degree in Chemical Engineering from the Georgia Institute of Technology and Honorary Doctorate in Physics from International Solomon University. Mr. Honeycutt is married to Margaret Lisette Schmidt. They have three children and nine grandchildren.

ABSTRACT

It is well known that liquid natural rubber latex (NRL) from the *Hevea brasiliensis* tree contains a number of proteins that cause minor to severe allergic reactions. Estimates run as high as 17% of healthcare workers affected by various types of latex allergies that have been studied extensively.[1,2] This paper proposes a unique method to alleviate the impact of normal levels of antigenic proteins but not negatively impact the needed chemical and physical properties. Past attempts at fixing the antigenic protein (AP) problem have usually resulted in a reduction in the chemical and physical properties of NRL. These authors believe that the natural protein plays an important role in the physical and chemical properties of NRL and AP reduction techniques that degrade the proteins are deleterious to the NRL properties. The key property of NRL is its elastomeric behavior and any alterations to that property reduce its utility. Vystar Corporation has developed a novel, patented method for the deactivation of the AP in liquid NRL that involves the denaturing of these proteins. This new, patented method for fixing the protein problem results in equal or slightly improved chemical and physical properties in NRL thus allowing a seamless transition for manufacturers using the current commercially available product. Of particular interest is that the low antigenic NRL appears to have a slightly reduced initial modulus when compared to untreated NRL. A reduced initial modulus translates into greatly reduced fatigue for items such as surgeon's gloves or examination gloves that are worn for a considerable period of time. Barrier property studies are in the early stages, but proceeding as planned.

INTRODUCTION

Although latex as a protectant has a long history of usage dating back to the 1800s, the use of barrier goods like gloves and condoms increased tremendously in the 1980s. Due to its complex mechanical and chemical properties, latex is considered to have the best broad range of desirable properties. Despite significant development of synthetic rubber latex (SRL), natural rubber latex (NRL) consumption reached 1.08 million tonnes in 2004 and continues to increase.[3] The market share for dipped products in this consumption is 65%.[4] The primary considerations for this preference are economical and technical. The price of oil (main raw material for SRL) has increased significantly. Furthermore, the properties of NRL are superior when compared to SRL (for example, stability against environmental degradation).[5]

Many efforts have been made to remove proteins from NRL by physical and/or chemical methods: centrifugation, use of proteolytic enzymes and surfactants, leaching (including ultrasonic), irradiation by cobalt, using a strong chlorine or acid/hypochlorite wash (Clorox), use of other latex producing plants not containing the hevea type protein, identified as allergenic.[6-8] Most of these methods are complex and incomplete. These methods generally offer limited removal of allergenic proteins while the resultant latex product(s) may have diminished physical properties. Vystar has developed a relatively simple technological method for the removal of the antigenic proteins by treating NRL in the liquid phase with selected inorganic chemicals (including $Al(OH)_3$ and SiO_2 as alkali solutions) prior to vulcanization.[9,10] Vystar has named and trademarked the resultant natural rubber latex, Vytex™. These methods decrease the level of antigenic protein in latex articles down to ≤0.2_g/g. For successful commercialization of this process, it is necessary to understand the influence the additives have on the technological characteristics of NRL, quality of NRL with anti-protein additives and quality of products made from low-protein latex. Data on this subject (mainly pertinent to NRL with SiO_2 in the form of nano-particles or water dispersion) is available in the literature.[11-14] However, it is obvious that this data is lacking enough evidence for the successful development of a new low-protein NRL and resultant products.

The aim of the current work is to investigate the influence of anti-protein additives on the physical properties of NRL. This investigation will examine the various procedures used in industrial manufacturing of NRL. The concentration of anti-protein additives specified hereafter provides effective antigenic protein removal.[9,10]

EXPERIMENTAL

Treated latex film samples for tensile testing were prepared at Vystar's research laboratory (Reactive Energy – Canada) and at selected industrial latex companies. Table 1 displays the parameters for sample preparation. Vystar is working with three manufacturers of finished NRL products in several industries who shall remain confidential at this time and are identified as Company A, Company B, or Company C.

Table 1. Conditions for sample preparation

#	Company	Supplier of NRL and country - producer	Mode of production	Type of sample
1.	Reactive Energy – Canada	Centrotrade- combined	Batch	Fingers
2.	Company A	Centrotrade- combined	Batch	Fingers
3.	Company B	Firestone – Liberia	Continuous	Gloves
4.	Company C	NA – Thailand	Continuous	Gloves

Anti-protein additives were prepared from the powders of $Al(OH)_3$ (57% of Al_2O_3), and SiO_2 (99.8%, Fumed Silica, 0.011 micron) supplied by Sigma-Aldrich. The powders were dissolved in KOH (concentration of alkali was 50%) at room temperature in calculated ratios to provide an optimum concentration of anti-protein additives, and acceptable technological properties for dipping. The mixtures of NRL with anti-protein additives were agitated for 24 hours (SiO_2) and 72 hours ($Al(OH)_3$). The samples were compounded by standard additives, which are commonly used to produce latex goods. Typical additives and concentrations are illustrated in Table 2. Prior to dipping, the mixtures were agitated for ≥48 hours at room temperature in a covered vessel Reactive Energy, Company A and Company B laboratories). Three hours at temperature 50^0C + 16 hours at room temperature was the compound procedure used at Company C. Dipping was repeated (straight double dip) at Reactive Energy and Company A laboratories using glass or metal forms. Next, this procedure was followed by subjecting each dipped sample to temperatures ranging from 120 - 130^0C for 5 minutes. The dipped samples were vulcanized at the same temperature for 40 minutes and were leached for 1 – 2 minutes in boiling distilled water. Finally, the samples were detached from their formers, coated with cornstarch and dried at room temperature. Coagulated dipped samples produced at Company B and Company C were prepared slightly different. Warm formers (temperature 70 – 80^0C) were dipped into a solution of coagulant ($CaCl_2$ or $Ca(NO_3)_2$ and dried (temperature 90 – 95^0C). This was followed by a single dip (Coagulate dip) into the latex mixture. Drying (80 -100^0C), water leaching (50 – 90^0C), curing (100 – 140^0C), post curing leaching and powdering with cornstarch powder were prepared in accordance with procedures used by Company B & Company C.

Table 2. Compounding additives

Additive	PHR	Additive	PHR
Potassium hydroxide[c]	**0.05**	2-mercaptobenzothiazole, zinc salt	**0.25**
Sodium polynaphtalenesulfonate	**1.00**	Zinc oxide	**0.70**
Sulfur	**0.70**	Butylated hydroxytoluene	**0.50**
Zinc dimethyldithiocarbamate	**0.45**		

[c] KOH was added only for control (blank) samples (without our additives).

Properties for liquid NRL were controlled before and after compounding as following:

- pH – by a digital pH-meter;
- Total Solids, (TS, %), - by baking at temperature 130^0C and evaluation of residual weight of sample;
- Mechanical Stability (MST, sec.) @ 61.5 – 62.5% TS - by Hamilton Beach Mixer;
- Viscosity (_, cP) – by Brookfield viscometer;
- Chemical stability (Zinc Oxide Viscosity (ZOV), at 60 min) @ 57% TS - by Brookfield viscometer;
- Pre-cure – Chloroform Number (CN) – by mixing of latex with chloroform;
- Swollen Diameter (D_s, mm) – at treating latex circle (**ø** 38mm) with toluene.

All mixtures prepared for dipping (except samples made at Company C) had pH 10.8 – 11.2, TS 50 – 55% and _ 50 – 100cps. Gloves made at Company C were produced from the latex mixture with TS 34%.

For tensile measurements, ring samples (diameter 60 – 65mm and width 20mm) were tested for tensile measurements. The ring samples were produced by cutting off a finger of the glove sample or another wide part of a glove. The sample thickness was evaluated by using calipers (error ≤0.02mm) and width and length were measured using a ruler (error ≤0.5mm). The ring samples were tested using a customized tensile machine, which was designed and built specifically for Vystar. This machine provides a stepped deformation with measurement of force by digital gauge EXTECH model 475044 (error ≤10g) at increasing and decreasing of stress. The decreasing of stress test was performed without destruction of the sample. Decreasing of stress started when elongation reached ~ 80% from maximum elongation at break.

Identical films were compared using the testing machine previously described to perform (ASTM D 412- *Tensile Strength of Rubber, Elastomer Tensile)*. Test results demonstrate the difference in values of strength and expansion at break not exceeding 10%. For each NRL sample (control or latex with anti-protein additives), ≥4 samples were tested. Tensile tests were performed five days after the sample was created and exposed to room temperature for a relaxation of the stresses arising in a film during vulcanization.

Degradation of latex after exposure to temperature of 70^0C during 7 days (ASTM D3578- *Standard Specification for Rubber Examination Gloves*) was also evaluated.

Gloves produced at Company C were evaluated for leakage at Nelson Laboratories (USA). This test determines the barrier protection confidence level (relative number of allowable defects – pinholes). The test procedure was performed according to (ASTM D5151-92- *Standard Test Method Detection of Holes in Medical Gloves* (3.1) and USFDA in Federal Register, 800.20 pp.7-10. The procedure included securing a test glove to a plastic cylinder and filling the glove with 1000ml of water. The gloves were suspended and examined for visual leaks immediately after the addition of water and re-examined 2 minutes later.

RESULTS AND DISCUSSION

The Glove leakage test results (Table 3) indicate that gloves made with anti-protein additives had better liquid barrier protection compared to "untreated" latex gloves.

Table 3. Glove Leakage Test

Results for gloves produced from: Marking: "**0**" – pass; "**+**" – fail (**2** = 2 min., **V** – visible, **I** – immediate)																							
Vytex NRL Pilot Line						Company C Latex in Pilot Line (blank)								Vytex NRL in Laboratory									
Size 7			Size 7 1/2			Size 7				Size 7 1/2				Size 7					Size 7 1/2				
0	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**+V**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**

A comprehensive examination of physical properties was performed for treated and non-treated latex samples produced at Company A laboratory. Table 4 displays the results of these tests. Test data indicates that the majority of technological parameters for NRL with anti-protein additives are comparable to the controls (non treated) with some improvement. A modest deviation from industry requirements was noticed in a sample, which contained 0.10 wt% SiO_2. This deviation was corrected by reducing the concentration of SiO_2 to 0.02 wt%.

Table 4. Results of technological tests for Vytex natural rubber latex at pilot trial

Property	Company A Industry range	NRL – Control “Blank”	NRL + 0.15 wt.% $Al(OH)_3$	NRL + SiO_2 0.10 wt.%	NRL + SiO_2 0.02 wt.%
Raw latex testing					
TS, %	61.0 – 61.5	**61.5**	**61.3**	**61.5**	**61.5**
pH	10.2 – 10.8	**10.82**	**11.02**	**10.98**	**10.79**
MST, sec	250 - 750	**594**	**1100**	**1679**	**600**
_, cP	40 - 90	**80**	**108.5**	**87.5**	**75**
ZOV, sec. @5 min.		**60**	**72**	**50**	**40**
ZOV, sec. @60 min.	80 - 600	**196**	**228**	**60**	**90**
Compounded latex testing					
TS, %	50.0 – 55.0	**51.2**	**50.9**	**50.8**	**55**
pH	10.8 – 11.2	**10.70**	**10.62**	**10.89**	**10.99**
MST, sec	-	**294**	**1492**	**1500+**	**750**
_, cP	-	**20.5**	**58.5**	**27.0**	**45**
CN	2+ - 4	**2**	**2**	**3**	**2**
D_s, mm	65 - 70	**70**	**70**	**70**	**70**

The technological properties can be influenced with anti-protein and compounded additives, which were examined during this study. Adjustments can be made by diluting the latex sample with water and/or potassium hydroxide after compounding or prior to adding the anti-protein additives. Table 5 illustrates an example of adjusting the latex sample after compounding at a recent trial performed at Company B.

Table 5. Technological properties for Vytex natural rubber latex at pilot trial at Company B

Properties	NRL – control blank	NRL + 0.15 wt.% $Al(OH)_3$	NRL + 0.02 wt.% SiO_2
After mixing with anti-protein and compounded additives			
TS, %	**58**	**58**	**58**
pH	**10.10**	**10.41**	**10.59**
_, cP	**195**	**975**	**445**
After adjustment before production of gloves			
TS, %	**55.3**	**50.2**	**52.5**
pH	**10.90**	**10.98**	**11.03**
_, cP	**90**	**72**	**53**

Table 6 illustrates adjustments made by adding diluted solutions with anti-protein additives to latex mixtures prepared at Vystar’s laboratory for samples used for tensile measurements.

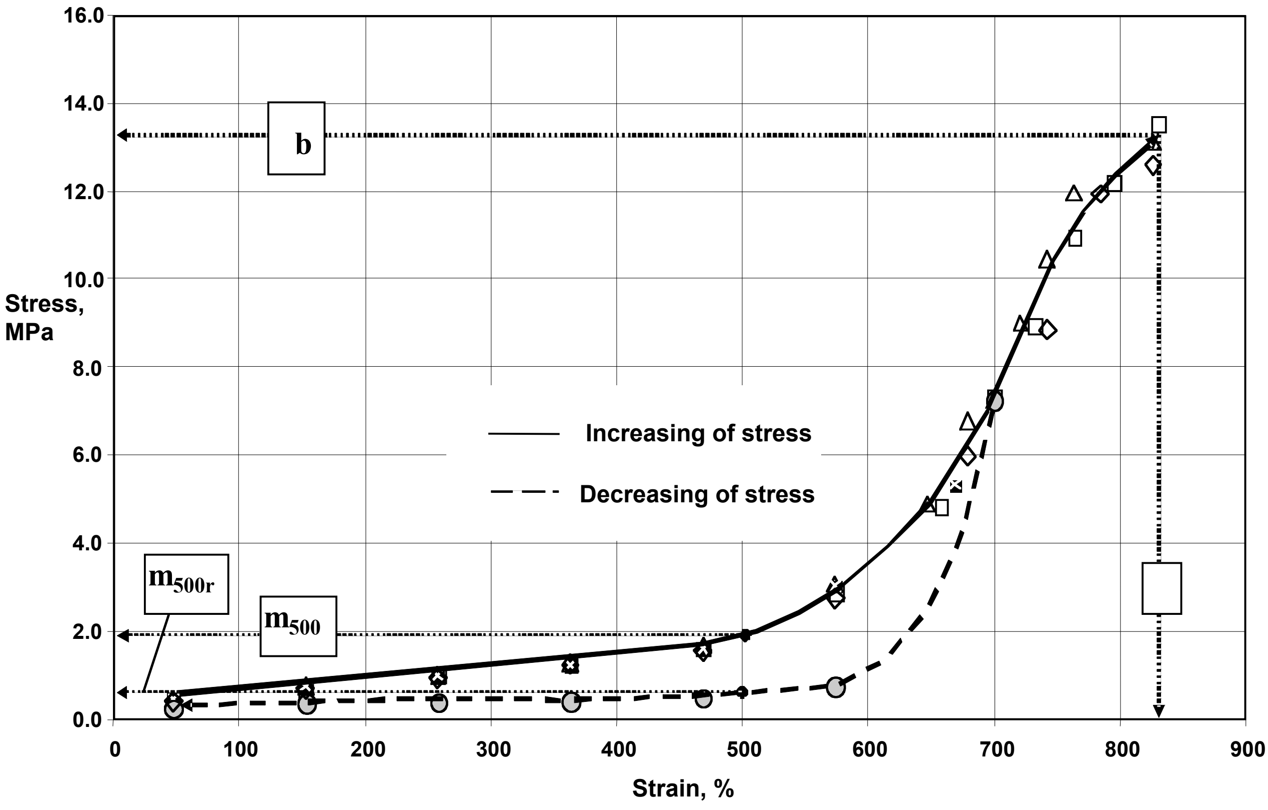
Table 6. Technological properties of Vytex natural rubber latex at different concentrations of anti-protein additives

Properties	NRL – control blank	Concentration of $Al(OH)_3$ in NRL (wt.%)	Concentration of SiO_2 in NRL (wt.%)

		0.10	0.15	0.20	0.02	0.05	0.08
Before mixing with compounded set (after mixing with anti-protein additives)							
TS, %	**-**	**-**	**-**	**-**	**-**	**-**	**-**
pH	**10.91**	**50**	**50**	**42**	**60**	**55**	**85**
_, cP	**82**	**10.88**	**11.13**	**10.94**	**11.04**	**10.94**	**11.03**
After mixing with compounded additives (before production of fingers)							
TS, %	**57**	**55.7**	**53.3**	**57.2**	**57.5**	**54.8**	**57**
pH	**11.02**	**11.18**	**11.30**	**11.15**	**11.00**	**10.97**	**10.82**
_, cP	**100**	**70**	**80**	**50**	**80**	**55**	**120**

Figure 1 exhibits a typical Stress-Strain diagram for the untreated samples. The following indices, which are listed below and illustrated in Fig. 1, are used for material characterization and comparison:

Figure 1. Stress-Strain curve for untreated latex



- $\_{b}$ - strength at break;
- _ - elongation at break;
- **m_{500}** – modulus @ deformation 500% at increasing of the stress;
- **m_{500R}** – modulus @ deformation 500% at decreasing of the stress;

- $\_{R}$ - residual (plastic) elongation by evaluation of changing of distance between marks on the sample after test;
- **m_{500}/m_{500R}** – ratio, which describe the loss of the strength by material at deformation.

Tables 7 - 10 present results of the tests at stretching of the latex rings.

Table 7. Results of tensile tests for Vytex natural rubber latex using straight dip process at pilot trial at Company A

Description of samples		Tensile parameters		
		_$_{b}$, MPa	_, %	m_{500}, MPa
NRL – control blank	After production of films	**16.5**	**851**	**1.8**
	After aging (7days@70^{0}C)	**13.3**	**684**	**2.7**
NRL + 0.15 wt.% $Al(OH)_3$	After production of films	**18.1**	**860**	**1.8**
	After aging (7days@70^{0}C)	**18.5**	**825**	**2.2**
NRL + 0.10 wt.% SiO_2	After production of films	**11.1**	**932**	**1.1**
	After aging (7days@70^{0}C)	**12.4**	**955**	**1.1**

Degradation of the blank sample after aging confirms the literature data[3]. However, latex, which was treated with anti-protein additives, demonstrates a better resistance to aging. This statement remains valid even for the latex containing additional quantities of silicon oxide (0.10 wt.%). Table 7 demonstrates that latex treated with SiO_2 leads to a decrease of strength at break point, however stability against degradation remained high.

For this composition, it would be necessary to adjust the vulcanization environment to keep the properties in required limits.

Table 8. Results of tensile tests for Vytex natural rubber latex at pilot trial at Company B

Description of samples	Tensile parameters			
	_$_{b}$, MPa	_, %	m_{500}, MPa	_$_{R}$, %
NRL – control blank	**14.7**	**740**	**4.0**	**10**
NRL + 0.15 wt.% $Al(OH)_3$	**17.7**	**720**	**4.3**	**10**
NRL + 0.02 wt.% SiO_2	**17.8**	**730**	**4.4**	**10**

Tensile strength of the latex sample (gloves) was increased ~20% after treatment with anti-protein additives during the pilot trials at Company B.

Table 9. Results of tensile tests for Vytex natural rubber latex at pilot trial at Company C

Description of samples	Tensile parameters					
	_$_{b}$, MPa	_, %	m_{500}, MPa	_$_{R}$, %	m_{500R}, MPa	m_{500}/m_{500R}
NRL – control blank	**15.4**	**840**	**2.0**	**7.5**	**0.28**	**7.1**
NRL + 0.04 wt.% SiO_2	**15.8**	**900**	**1.9**	**7.5**	**0.30**	**6.3**

Tensile strength and ultimate elongation of the latex sample (gloves) were increased and modulus slightly decreased after treatment with anti-protein additives at the pilot trials at Company C.

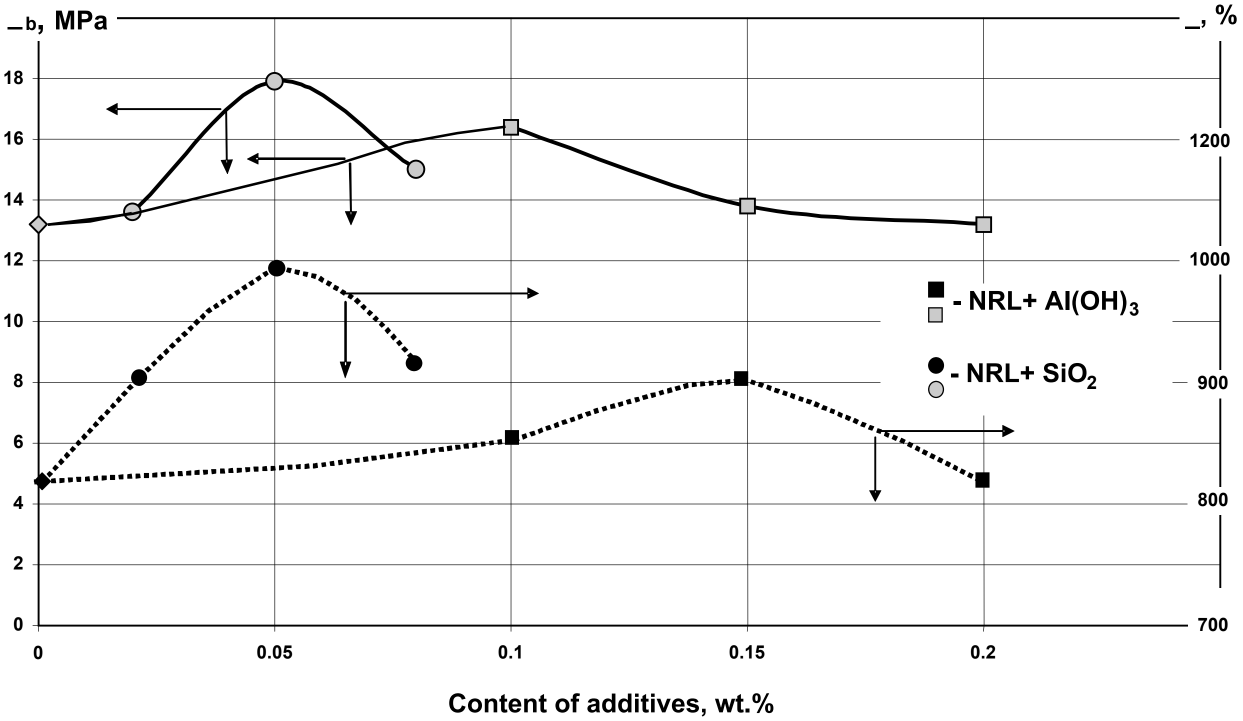
Table 10. Tensile properties of NRL at different concentration of anti-protein additives (Vystar laboratory)

Description of samples	Tensile parameters					
	_$_{b}$, MPa	_, %	m_{500}, MPa	_$_{R}$, %	m_{500R}, MPa	m_{500}/m_{500R}
NRL – control blank	**13.2**	**820**	**2**	**11**	**0.30**	**6.7**
NRL + 0.02 wt.% SiO_2	**13.6**	**900**	**1.6**	**11**	**0.25**	**6.4**
NRL + 0.05 wt.% SiO_2	**17.9**	**980**	**1.5**	**13**	**0.4**	**3.8**
NRL + 0.08 wt.% SiO_2	**15.0**	**910**	**1.6**	**12**	**0.35**	**4.6**
NRL + 0.10 wt.% $Al(OH)_3$	**16.4**	**850**	**2.0**	**10**	**0.33**	**3.3**
NRL + 0.15 wt.% $Al(OH)_3$	**13.8**	**900**	**2.0**	**11**	**0.50**	**4.0**
NRL + 0.20 wt.% $Al(OH)_3$	**13.2**	**820**	**1.8**	**8**	**0.30**	**6.0**

Based on the data from Tables 3, 7-10 it is possible to conclude that a defined concentration of anti-protein additives is required to maintain the valuable physical properties of products made from NRL. Some parameters, such as tensile strength, ultimate elongation, and resistance against aging are increased when the amount of anti-protein additives in NRL was increased within a certain range. Understanding the relationship between anti-protein additives amounts and the properties of NRL is critical because exceeding the desired concentration amount will result in a deterioration of the mechanical properties of latex products.

Figure 2. illustrates this statement in the form of a diagram. It is obvious that mechanical properties of NRL with a small amount of the anti-protein additives are superior compared to NRL without these additives. Maximum values on the curves correspond to concentrations of the additives, which provide satisfactory technological parameters of liquid latex and an effective removal of the antigenic protein.

Figure 2. Mechanical Properties of latex films at different amount of anti-protein additives



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CONCLUSION

Vystar Corporation has developed a novel, patented method for the antigenic protein deactivation in barrier products (gloves, condoms) produced from natural rubber latex. This new method involves the treatment of liquid latex with special anti-protein additives, such as alkali solution of aluminum hydroxide or silicon oxide. The resultant latex products decreased in antigenic protein value to ≤0.2_g/g. Test data confirms the technological properties of the new low-protein latex can be adjusted to acceptable levels within the latex industry. It allows the production of high-quality products from new low-protein latex without changing existing technology. Anti-protein additives increase the elasticity and strength of the latex films if concentrations of these additives are not exceeded. Removal of the antigenic protein occurs at sub-critical concentrations of anti-protein additives. Simultaneously, the barrier properties and resistance against aging of latex products with these additives are not only maintained, but also essentially improved. Vytex natural rubber latex has passed several successful pilot trials in the industry.

ACKNOWLEDGMENTS

The authors are grateful to the following colleagues for their contribution to this work: Isaac Maistrovski for his assistance in the laboratory routine; Russell Culp for his technical review and editing.

REFERENCES

1. Yassin MS, Lierl MB, Fisher TJ, et al. Latex allergy in hospital employees. Ann Allergy 1994;72:245-9
2. Kelly KI, Pearson ML, Kurup VP, et al. A cluster of anaphylactic reactions in spina bifida during general anesthesia. J Allergy Clin Immunol 1994;94:53-61.
3. Chen FB. Synthetic Polyisoprene Latex. Presented at the International Latex Conference, 2005; Charlotte, North Carolina.
4. Smit H. An Overview of Rubber Lattices. Presented at the International Latex Conference, 2005; Charlotte, North Carolina.
5. Office of Science and Technology Annual Report. Environmental Degradation of Latex Gloves: the Effects of Elevated Temperature on Tensile Strength. Food and Drug Administration, 1996.(16):2-4.
6. Removal of Protein from Natural Rubber Latex. JETRO 1994;10,001-05.
7. Eng A, Kodama S, Kawasaki H. Reduction of Water Extractable Proteins in Natural Rubber Latex Films by Ultrasonic Leaching System. Journal of Rubber Research 1999;2(1):23-26.
8. Cornish K, Williams J, Martin JA. Commercialization of Yulex Natural Latex™: the improving margin structure of guayule latex products. Presented at the International Latex Conference, 2005; Charlotte, North Carolina.
9. Honeycutt T, et al. Decreasing allergenicity of natural latex rubber prior to vulcanization, US Patent Application No 11/096,421 filed October 12, 2005.
10. Honeycutt T, Sharivker V, Sharivker S, Blinov V, Doyle W. A Novel “Protein-Free” Natural Rubber Latex: Properties and Applications. Presented at the International Latex Conference, 2005; Charlotte, North Carolina.
11. Amdur S, et al. Removing Natural Latex Proteins from Dipped Rubber Goods with Fumed Silica Additives. Presentted at the meeting of the Rubber Division, American Chemical Society, Orlando, Florida. Published by SMTL, February, 2000 Version 1.0.
12. Amdur S, et al. Latex compositions and articles manufactured therefrom, US Patent 5,458,588 A61F 13/15, July 19 1988.
13. Amdur S, et al. Viscosity enhanced natural rubber latex dipping media and dipped articles of greater thickness prepared therefrom, US Patent Application 20020156159 A1, March 15, 2002.
14. Xiong M, et al. Preparation and Characterization of Acrylic Latex/Nano-SiO_2 Composites. Polymer International, V. 51, #8, 2002;693-698.