Materials Research

REFERÊNCIA

RIBEIRO, Jaqueline Alves et al. Development assessment of natural latex membranes: a new proposal for the treatment of amblyopia. **Materials Research**, São Carlos, v. 20, n. 3, p. 653-660, maio/jun. 2017. Disponível em: <http://www.scielo.br/scielo.php?script=sci_arttext&pid=S1516-14392017000300653&lng=en&nrm=iso>. Acesso em: 18 jan. 2018. Epub Mar 30, 2017. doi: http://dx.doi.org/10.1590/1980-5373-mr-2016-0355.

Materials Research. 2017; 20(3): 653-660
DOI: http://dx.doi.org/10.1590/1980-5373-MR-2016-0355

Development Assessment of Natural Latex Membranes: A New Proposal for the Treatment of Amblyopia

***Jaqueline Alves Ribeiro*[a,b]*, Suelia Rodrigues Fleury Rosa*[b], Cicilia Raquel Maia Leite*[c], *Claudio Lopes Vasconcelos*[d], *João Maria Soares*[d]**

[a] *Instituto Federal Goiano, Campus Ceres, GO, Brasil*
[b] *Laboratório de Engenharia e Biomaterial - BioEngLab®, Faculdade Gama, Universidade de Brasília, Brasília, DF, Brasil*
[c] *Laboratório de Engenharia de Software - LES, Ciência da Computação, Universidade do Estado do Rio Grande do Norte, Mossoró, RN, Brasil*
[d] *Laboratório de Análises Magnéticas e Ópticas - Lamop, Universidade do Estado do Rio Grande do Norte, Mossoró, RN, Brasil*

Received: May 05, 2016; Revised: October 25, 2016; Accepted: March 03, 2017

The ophthalmic dysfunction amblyopia, commonly known as lazy eye, is characterized by decreased vision in one eye due to improper development in childhood. The aim of this study was to obtain and characterize natural rubber membranes and to assess their utility as an eye film capable of altering the passage of light. The latex membranes were produced using the *Van Gogh* method and the deposition technique and were analyzed by physical and chemical methods to determine the properties of latex *in natura* and of natural rubber membranes. The materials were characterized by X-ray diffractometry, scanning electron microscopy, thermogravimetry, differential scanning calorimetry, analysis of water sorption and light crossing analysis. We report here a new approach to the treatment of patients with amblyopia using latex membranes.

Keywords: *Membranes; Amblyopia; Latex; Biomaterial*

1. Introduction

Amblyopia is an eye pathology that develops during childhood. It is characterized by decreased visual acuity in one eye, which can be attributed to abnormal visual development during the early years of life, and few treatments are available. The prevalence of amblyopia in children is estimated to be 1-4% around the world, and it is the most common cause for the decrease in visual acuity in childhood[1]. One of the methods used for the treatment of amblyopia is optical correction followed by occlusion of the eye with better visual acuity to enable the weaker eye to develop[2]. Research on the effects of occlusion has shown that secondary deprivation effects may arise from fixation of the eye and that the occlusion time should not be longer than 6 hours[3]. This trend has also been observed in the ophthalmic community in Brazil, although there is a lack of national clinical evidence[4].

Rapid technological development and advances in medicine have substantially improved the life quality of people. One of the advancement is the development of biomaterials, which are used to replace, partially or totally, the lost or damaged organic tissues. Besides the biocompatibility, suitable biomaterials for use in medical devices should be easily molded into complex shapes. Generally, biomaterial's requirements can be grouped into four broad categories: biocompatibility, ability to be sterilized, functionality, and reproducibility[5].

Biomaterials can be used to manufacture contact lenses[1], since they are defined as active substances capable of interaction with the surrounding tissue without causing an immune response[6]. Natural latex from the rubber tree, *Hevea brasiliensis,* is a raw material of high quality and durability, with biocompatible physical and chemical characteristics, antigenicity, hypoallergenicity, impermeability, elasticity, softness, flexibility, and low cost and resistance. These characteristics are in agreement with the most current scientific studies and provide patients with increased comfort and reduced risk of developing allergies.

It should be noted that latex has already been used for the production of esophageal prostheses, biomembranes, and an esophageal flow controller module[7]. Latex is excellent for inducing repairs, such as in bone loss and injuries to skin and blood vessels[8]. *In vivo* tests in the conjunctiva of rabbits showed that natural rubber membranes promote healing of conjunctival scarring and angiogenesis[9]. Andrade et al.[10] study was carried out to compare the immunobiological mechanisms and the oxidative stress status involved in tissue healing stimulated with these membranes. In this study, we chose the latex, LENCOC®, as the material for the production of membranes based on previous research and because it is a simple material that is easy to handle[11,12].

* e-mail: jackalvesribeiro@gmail.com

Latex is a polyphasic and polydisperse colloidal system containing poly (*cis*) isoprene molecules in suspension. The latex stability is due to the presence of water-insoluble proteins and lipids that act as natural surfactants. Natural rubber latex comprises particles of rubber hydrocarbon, which are non-gummy and suspended in an aqueous serum phase whose average composition is 36% oil, 1.4% protein, 1.6% carbohydrates, 1% neutral lipids, 0.6% glycolipids and phospholipids, 0.5% inorganic components, 58.5% water, and 0.4% other substances[7].

The physiological functions of latex in the plant are: transport and storage of nutrients; providing protection to the plant through promoting wound healing and controlling insect attacks; as a water reserve during dry seasons; and providing continuous tissues supplies derived from cyclopentose via the cyclic pentose phosphate pathway, which is essential for development[13]. The methods used for the production of latex-based devices have been used since 1996 when it was first proposed and described by Mrué[14], cited in[11] who developed a production method that has been validated to date for numerous artifacts, to be used as inducers of neovascularization and tissue repair.

However, this traditional method cannot be applied to the manufacture of occlusion lenses because of the high density of the chemically heterogeneous latex and the presence of impurities. Therefore, we focused on techniques that allow the degradation of polymers, and, if possible, the regeneration of the monomers for later use without introducing hydroxide and formaldehyde. Thereby, latex will be as a translucent rubber film that will allow light to pass through in irregular paths (and thus prevent clear visualization of the object), with diffuse reflection and minimum absorption of light to prevent heat generation.

We present here a simple approach to eye occlusion based on an ophthalmic membrane made of an occluding biomaterial derived from natural rubber latex. Natural rubber membranes were developed using two different techniques, the *Van Gogh* technique and the deposition technique. The different membranes were characterized by physical and chemical methods to determine the properties of the latex *in natura* and of the membranes subjected to heat treatment at 40ºC and 70ºC. The materials were characterized using X-ray diffractometry (XRD), scanning electron microscopy (SEM), thermogravimetry (TG), differential scanning calorimetry (DSC), and analysis of water sorption and light crossing analysis. Based on their physical and chemical properties, the applicability of latex membranes from natural rubber as biocompatible ophthalmic occluders was evaluated.

2 Materials and Methods

2.1. Preparation of the raw material

The latex used was purchased from the domestic market. It had already gone through the centrifugation process and contains suspensions of sulfur and resin purchased from the suppliers in the Southeast region. High sulfur concentration gives the latex, after vulcanization, an adhesive property and low viscosity. However, it would require a long time to conclude the development of the membranes, therefore, was chosen the double centrifugation and pre-vulcanization latex.

From the natural latex, a final compound was prepared by the addition of chemicals by following the steps previously described[11]. This procedure provides the latex membrane (LENCOC®) essential characteristics for a lens-elasticity, softness, strength, impermeability, and hypoallergenicity.

2.2. Preparation of the mold

The technique development to obtain the latex membrane was performed in the Biomaterial Engineering Laboratory - BioEngLab®, at the Faculdade UnB Gama, according to the steps for the production of the mold and membrane, as described below.

In the *Van Gogh* technique, a mold has been developed. It was made of plastic material coated with an acrylic layer, measuring 3 cm in width, 3 cm in length, and 1.5 cm in diameter as previously described[11]. The production of the membrane need be customized for each patient, in order that its shape and proportions to follow the characteristics of the patient's eyes, thus providing greater comfort.

2.3. Preparation of the membranes

The *Van Gogh* technique was based on the immersion technique[14,15], which consists on the insertion of the molds into the final latex compound, perpendicular to the plane, in a gradual and uniform way, followed by heating in an incubator with a thermostat. This step represents the beginning of polymerization, which determines the final characteristics of the product. After this phase, the mold was removed slowly and gradually, placed in the incubator and subjected to vulcanization at a temperature of 40ºC. The molds were then removed and the membranes were prepared using thick brush strokes by means of a flexible rod with cotton-covered tips, 1 cm away from the mold, moving back and forth to completely cover the mold. This procedure was repeated by three or four times in succession, followed by drying in an incubator at temperature of 40ºC, with 2-hour time intervals in order to obtain membranes with a thickness up to 0.2 mm. After this drying period, the *Van Gogh* membranes were maintained in their state of rest for 24 hours at room temperature to complete the procedure.

The membranes were obtained by deposition on the petri dish as described previously[16,17,18]. The transparency of the membrane was favored by the use of room temperature. The vulcanization procedure can leave the membrane with a more yellow appearance. However, for the preparation of these membranes, an average of 3 to 4 days for total drying and polymerization are required, because the structure

and shape of the membrane must be uniformly preserved; thus, a more thorough preparation process is needed. The membranes obtained by deposition in the petri dishes had a diameter of 8 cm and a thickness of 0.3 mm.

The latex membranes obtained by both techniques were developed using several different protocols to obtain a model that maintains the main features-occlusion of light, appropriate thickness, and applicability. The latex membrane will henceforth be identified as *Van Gogh* membranes obtained from three baths (M1) and four baths (M2), both vulcanized at 40ºC; the membranes obtained by the deposition technique vulcanized at 40ºC will be referred to as M3 and those vulcanized at 70ºC will be referred to as M4.

The development of membranes started from the preparation of the environment and the raw material until the physical and chemical characterization of the membranes, which comprised a series of steps as presented in the flowchart in Figure 1.

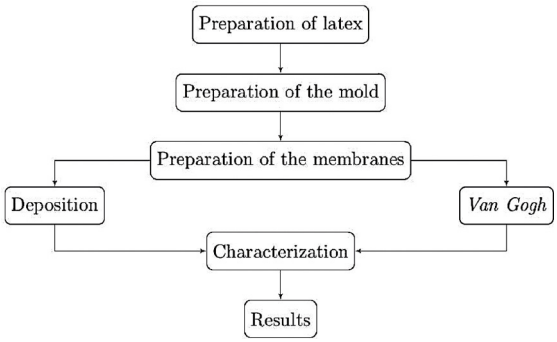

Figure 1. Flowchart of the stages of preparation and characterization of the membranes.

2.4. Characterization of the membranes

The prepared latex membranes M1, M2, M3, and M4 were characterized by X-ray diffraction, SEM, thermal gravimetric analysis (TGA), DSC, X-ray fluorescence analysis (EDX), water sorption analysis and light crossing analysis to investigate the characteristics of the latex *in natura* and of natural rubber membranes heat-treated at 40ºC and 70ºC. The membranes were characterized in the "Laboratory for Magnetic and Optical Analyses" and in the "Laboratory of Catalysis", Environment and Materials, both at the State University of Rio Grande do Norte.

The fluorescence was measured at room temperature on a fluorescence spectrophotometer (SHIMADZU EDX-7000, Energy Dispersive X-Ray Fluorescence Spectrometer).

XRD (Rigaku Miniflex II RV-200B X-ray diffractometer) of latex membranes was performed using a Cu-Kα target at 30 kV and 30 mA. The diffraction angle was varied within the range of $5º < 2\theta < 50º$ at a scan rate of 2º/min.

SEM images of the latex membranes were obtained using a TESCAN MIRA 3 FFG electron microscope (filament, tungsten; voltage, 25.0 kV). Prior to the analysis, all samples were covered by a carbon layer and metalized with gold.

About the TGA, differential thermogravimetric (DTG) analysis, and DSC were performed using a Netzsch Leading Thermal Analysis STA 449 F3 Jupiter. All analyses were performed with a 5 mg sample in aluminum pans under a dynamic nitrogen atmosphere between 25ºC and 800ºC. These experiments were performed at a scanning rate of 10 K/min.

The water sorption measurements were performed by immersing the membranes in saline solution (0.9% NaCl). The ability of the membranes to absorb water salt solutions (aqueous solutions of 0.9% NaCl and 0.1% purified water; physiological saline) was determined using the initial dry mass as a parameter for monitoring the mass increase over time. Samples of vulcanized membranes with an area of 1 cm^2 were hydrated and kept in 25 ml of saline solution for 24 hours at room temperature and humidity throughout the duration of the experiment. At pre-set time intervals, the membranes were removed from the solution, dried on absorbent paper, and weighed quickly. This entire procedure was performed quickly, and at each weighing, the membranes were immersed in the saline solution again. At the end of the experiment, the water sorption capacity of each membrane was analyzed.

The variation in mass gain was calculated according to equation: $\frac{m_i - m_s}{m_i} \times 100$, where m_i is the initial mass and m_f is the final mass of the moist membrane.

About the Light crossing analysis, an optical device called focimeter was used. It focuses the optical system for which light is sent to evaluate the degree of the lens; the scale interval that appears on the display is a dioptric numerical value. For this characterization, a Nidek CLE-60 digital focimeter was used, in an optometry clinic in Goiânia, GO, Brazil.

To read the dioptric numbers, the sharpness of the reticles in the focimeter needs to be adjusted. The reticles are images overlapping the display that represent the light refracted in a lens of a certain diopter. When we make this reticle clear, it means diopter X of the lens being read is ideal for the formation of that particular image, and the dioptric readings are performed[19].

The light crossing analysis was performed with the objective of evaluating the passage of light trough the membranes and verifying the behavior of the surface light on the rugosity of the *Van Gogh* technique. It is worth mentioning that the equipment that was used for this analysis was perfectly calibrated so the measurements present a high degree of reliability.

3. Results and Discussion

About the Fluorescence measurements, the X-ray fluorescence technique (EDX) was used to identify the chemical components in the membrane after every process. As the amount of energy released is specific to each element, it is possible to identify the chemical elements that are

present at the point of incidence of the beam. In addition to identification of chemicals, this device also allows mapping of their distribution and generation of compositional maps for each desired element.

Table 1 presents EDX data obtained on the chemical composition of the membranes. High amounts of sulfur and potassium were detected on all membranes, and the quantities of phosphorus and zinc were lower than other components. The membranes developed using the *Van Gogh* technique, M1 and M2, had slightly higher quantities of sulfur than the membranes developed using the deposition technique. The M4 had lower amount of sulfur because of vulcanization at 70ºC, which increased the amount of potassium in relation to that in other membranes. This reaction occurred because the *Van Gogh* technique uses a smaller amount of the compound. M1 was thinner (3 baths) than M2 (4 baths) in the vulcanization process at 40ºC, suggesting that vulcanization is faster in a thinner layer.

Using EDX, we identified the components in the latex, in the preparation and centrifugation techniques according to the samples of natural rubber membranes, both belonging to isoprene, the main constituent of latex, as well as to the functional groups.

It's possible to see better solubility of sulfur than potassium. Therefore, when the membrane is vulcanized at high temperatures, there is higher molecular mobility; therefore, the amount of sulfur decreases and the amount of potassium increases, for M4 membrane. The sum of S and K remains roughly the same at all samples, accounting for more than 90 % of total samples constituent content, aside from carbon. Zinc had increased in M3 due to the slow vulcanization of the latex compound in the Petri dish.

Other metals, such as copper and iron, didn't have significant representation. These minerals will not cause allergy in contact with the eye, since they're already part of the human body and can also be found in food, which are mandatory for the function of biological systems.

The Figure 2 shows the X-ray diffraction profiles of M1 and M2, developed using the *Van Gogh* method, and M3 and M4, developed using the deposition technique. The X-ray diffraction profile of the membranes is predominantly amorphous and has a reflection band at $2\theta = 19º$, which is in agreement with data previously reported in the literature[20,21,22]. Although the membranes present an amorphous character, a notable difference between the diffractograms of the membranes can be observed depending on the preparation technique. The X-ray diffraction profiles of membranes M3 and M4, developed at different vulcanization temperatures, showed some crystallinity, which was different from that of the membranes M1 and M2. Notably, increasing time involved in preparing the membranes increased the chemical crosslinking between isoprene segments, in which explains the higher crystallinity shown by the latex membrane prepared using the deposition technique.

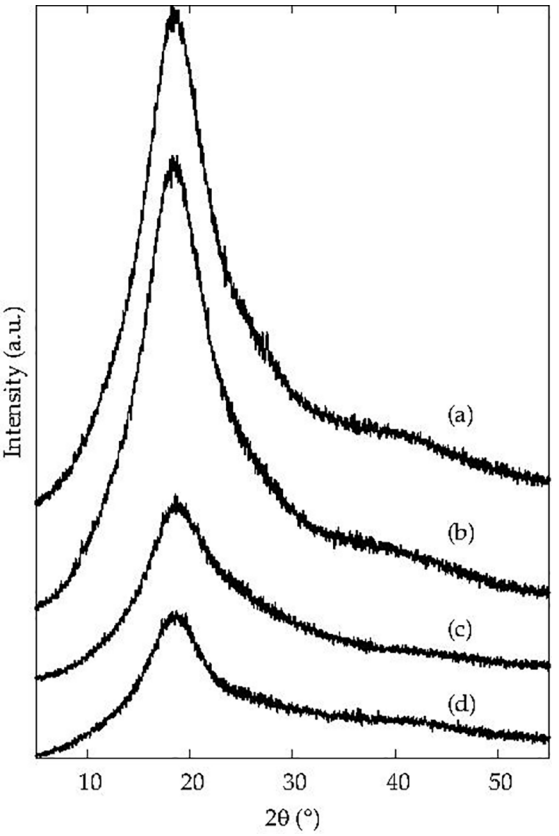

Figure 2. X-ray diffractograms of latex membranes M4 and M3 and M1 and M2 prepared using the deposition technique and Van Gogh method, respectively.

The Figure 3 shows the TG and DTG curves for the M1 and M2 *Van Gogh* membranes; for M3 and M4 membranes developed using the deposition method; and for the natural rubber latex (NRL). The complete degradation of sulfur occurs at a temperature of approximately 300 ºC, which is consistent with the onset of thermal degradation of the NRL and membranes. For all samples, no significant degradation was observed before 320 ºC. The maximum degradation temperature of the polymer was approximately 375 - 377 ºC, which demonstrates that the preparation techniques did not

Table 1. Chemical composition of the M1, M2, M3, and M4 membranes.

	S	K	P	Zn	Cu	Fe	Others
M1	79.70	16.80	2.20	1.00	0.17	0.08	0.05
M2	70.50	25.60	2.40	1.12	0.15	0.10	0.13
M3	69.70	21.18	1.45	6.36	0.49	0.22	0.60
M4	57.77	32.86	3.78	3.82	0.76	0.23	0.78

significantly affect the thermal behavior of the membranes. Thermogravimetric data obtained from TG and DTG curves are shown in Table 2. It's possible to observe that the temperature ranges in which can be observed the stages of degradation, mass losses established for each event, and final residue.

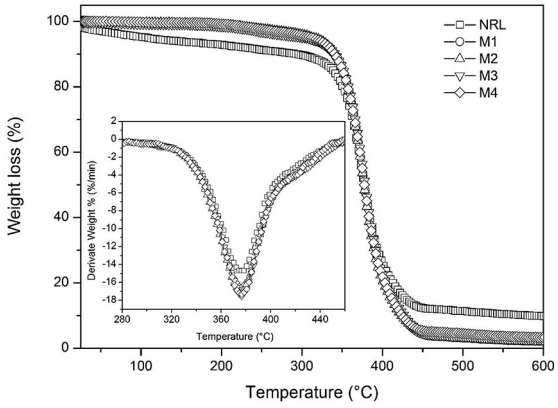

Figure 3. TG and DTG curves for natural rubber latex (NRL) and membranes M1, M2, M3 and M4.

Further inspection of thermal degradation of the membranes in Figure 3 shows that the DTG curves present a slight asymmetry evidenced by a slant around 400 - 460 ºC. This assymetry is due to the slow decomposition of polymer chains or highly cross-linked polymeric residues. Complete degradation of the membranes as well as that of the NRL occurred in a single stage, which was completed at a temperature range of 464 - 469 ºC. These results are similar to those found in the literature[23]. The mass loss observed in this temperature range can be linked to the oxidative pyrolysis of rubber, vaporization, and removal of volatile products. In thermodegradation of polyisoprene, fragmentation leads to depolymerization of the macromolecules generating the main products, isoprene and dipentene[24]. Above 470 ºC no other significant decomposition event was observed, suggesting the presence of only decomposed gas, ash, and inorganic species (oxides, carbonates, phosphates and metal silicates) at this stage, which could be due to the presence of impurities or substances that would be naturally present in latex.

The Table 2 shows T_0 as the initial temperature of thermal degradation; T_p as the maximum temperature degradation; T_f as the final temperature of that degradation stage; C_0, C_p, and C_f are the mass percentage in T_0, T_p, and T_f, respectively.

The Figure 4 shows the DSC curves for NRL, for the *Van Gogh* M1 and M2 membranes and for the M3 and M4 membranes prepared by the deposition method. Notably, all the membranes and NRL presented exothermic events.

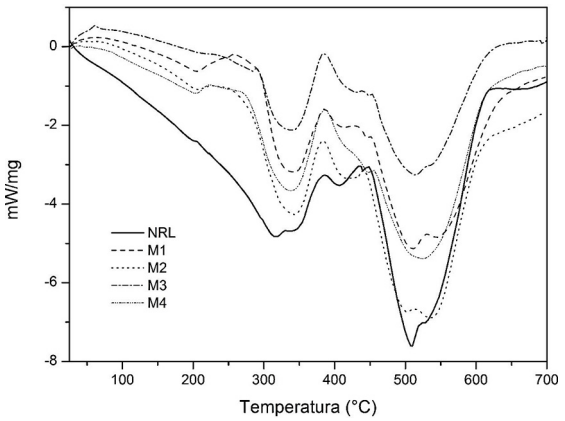

Figure 4. DSC curves for natural rubber latex (NRL) and membranes M1, M2, M3 and M4.

The thermal behavior of the latex membranes is similar to the NRL (Figure 4), where the exothermic processes are clearly observed. The exothermic event observed in at 350 - 460 ºC is consistent with the macromolecular thermodegradation of the membranes. First, the crosslinking between the polymer chains breaks in the vulcanization process. This is followed by depolymerization of the compound, which coincides with the thermogravimetric results already reported in the literature[24]. In addition, a significant difference between the DSC curves of the membranes was observed in this temperature range in which we perceive a wider range of energy related to the M3 and M4 membranes obtained by deposition, compared to the M1 and M2 *Van Gogh* membranes. This is consistent with the X-ray diffraction profile of the M3 and M4 membranes, which had showed the increased crosslinking between the isoprene chains. This increased crosslinking would consequently release a greater amount of energy following thermal degradation.

About the scanning electron microscope, the Figure 5 shows the micrograph of the M1 and M2 membranes produced

Table 2. Thermogravimetric data obtained from the treated membranes, where T_0 is the initial temperature of thermal degradation, T_p is the maximum temperature degradation, T_f is the final temperature of that degradation stage, and C_0, C_p, and C_f are the mass percentage in T_0, T_p, and T_f, respectively.

	T_0 (ºC)	T_p (ºC)	T_f (ºC)	C_0 (%)	C_p (%)	C_f (%)
NRL	296.7	375.1	460.9	89.9	53.7	12.0
M1	297.6	376.1	469.0	95.2	53.2	3.1
M2	298.9	375.7	468.6	96.2	52.5	4.6
M3	303.2	376.3	464.1	95.1	53.7	4.2
M4	303.2	376.6	463.9	95.1	51.9	5.0

by the *Van Gogh* technique, heat-treated at 40ºC. The M1 membrane prepared using 3 baths showed roughness and porosity throughout the area of the micrograph, with the pores distributed throughout the membrane and allowing water sorption. In the M2 membrane, prepared using 4 baths was denser, as evidenced by the presence of rubber particles. This increased density decreased the size (less than 1 µm) and distribution of the pores throughout the membrane.

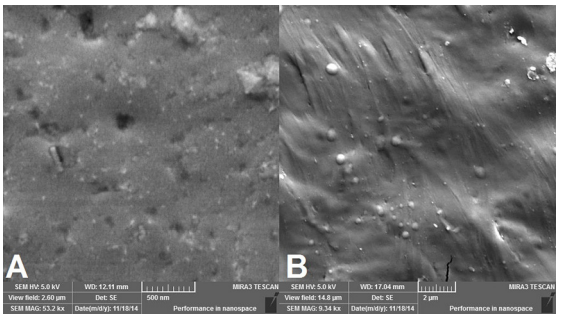

Figure 5. Micrograph of the Van Gogh membranes, (a) M1 and (b) M2.

Figure 6 shows the micrograph of the membranes produced by the deposition technique. M3, which was heat-treated at 40ºC, had showed the presence of rubber particles in a denser and more organized distribution. In M4, which was heat-treated at 70ºC, larger pores were observed.

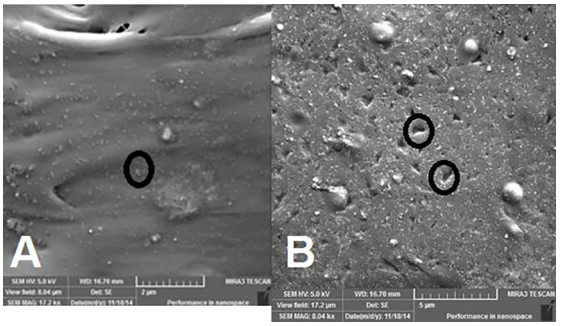

Figure 6. Micrograph of membranes produced by the deposition technique. (a) M3 and (b) M4.

The Table 3 shows the mass gain by the membranes and the time taken for stabilization. This analysis showed that M1 gained greater mass at a slower rate, while M4, which was heat-treated at 70ºC, stabilized in a shorter time. M3 absorbed the largest amount of water. M2 absorbed 4.4% water over a 105 min interval. Of the membranes produced at 40ºC, those produced by the 4-baths *Van Gogh* technique showed lower water absorption.

Talking about the light crossing analysis, the focimeter is used on specific materials for glasses and contact lenses. In an eyeglass lens, for example, the values presented in S and C represent the degree, with positive for astigmatism and negative for myopia; A represents the axis and the prism (Δ) the variance of the light beam.

Table 3. Absorption time of the latex membranes.

Sample	Time (minutes)	Mass
Membrane 1	375	5.4%
Membrane 2	105	4.4%
Membrane 3	105	9.0 %
Membrane 4	30	1.5%

In a normal contact lens, the values presented are: S = 0.25, C = 0.25, A = 24 and Δ = 0. These values represent two values for diopters and two values for the axis. When measuring the membranes, the numerical value of the instrument was disregarded. As such values cannot be assessed in latex.

In the focimeter device, has been observed the measurement of the light beam on the latex membrane. In this essay, five samples of each membrane were tested in order to verify their reproducibility. Figure 7 presents the assessment of the passage of light from membranes M1 (A), M2 (B), M3 (C) and M4 (D).

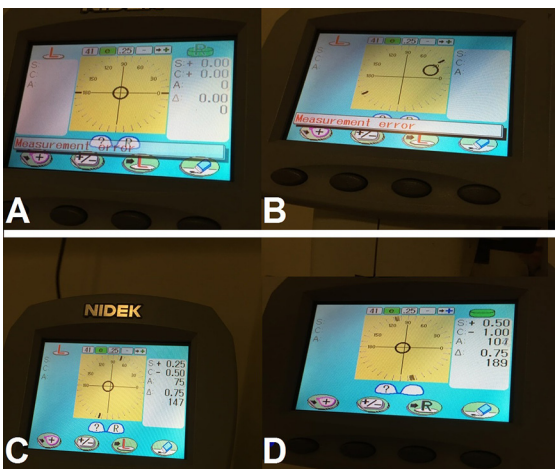

Figure 7. Measuring the membranes with the focimeter, M1 (a), M2 (b), M3 (c) e M4 (d). In the images, the angle of the focimeter display and the dark environment of the optometry clinic.

On membrane M1, could be observed that the beam locates the optical center, indicating a small passage of light. Due to the irregular surface caused by the brushes, the equipment shows an error in measurement. Consequently, the *Van Gogh* technique, with its brushes, made the surface rugose and gave a diffuse passage of light.

On membrane M2, all membranes did not present passage of light. The focimeter did not locate the optical center, which caused a measurement error. Membrane M2 has the most irregular surface due to a difference in manufacturing from M1, because the number of brushes was higher. This data prove the membrane is opaque, which is a characteristic of occlusion.

On membranes M3 and M4, produced with the deposition technique, the surface did not have irregularities. The focimeter performed the reading through the optical center, measuring the values, with no errors and total passage of light.

Thus, the *Van Gogh* technique can be considered as viable for the manufacturing of latex membranes for treating amblyopia. The passage of light was diffuse due to the rugosity and opacity, which will provide partial or total light occlusion in the eye of the patient. M1 presented a diffuse passage of light, which can stimulate the eye beyond the occlusion. M2 presented opacity with total occlusion of the light, which can benefit the treatment when compared with the eyepatch.

It is important to mention that the optometrist must investigate each case and identify the best treatment. With this, we aimed at using biotechnology and engineering to improve the therapy for a disease, that is related to visual disability, improving the quality of life of patients and thus reducing the possibility of future problems.

4. Conclusion

This optical engineering study highlights the potential use and great advantages of the LENCOC® latex membrane for a safe and comfortable option for treating amblyopia. During the characterization process, we defined four distinct types of membrane samples according to different temperatures, and the use of *Van Gogh* or deposition techniques. All of them have been tested using different equipment for the physical and chemical analysis of the biomaterials. The M1 membrane allowed the passage of diffuse light, causing partial occlusion, which preserves the movements and stimuli in the eye; Membrane M2 is opaque, without passage of light, and it is thin, transparent and has low water absorption; so, it can be used to treat amblyopia, replacing the cap with more comfort and providing a shorter treatment.

This study presented use of a LENCOC® latex membrane for the treatment of amblyopia. The *Van Gogh* technique demonstrated that the physical and chemical characteristics of the latex membrane present results compatible with the eye when applied as an occluder, reinforcing its biomaterial characteristic. The confection method is reproducible and the latex membrane is very stable.

Further *in vivo* studies are needed to determine the feasibility of this technique. The primary focus of this study was to characterize the latex for use as an occluder for the treatment of amblyopia.

Owing to the importance of biosafety in medical bioengineering research, extensive studies to confirm the efficacy of new treatment methods are critical. Thus, the development of natural latex membranes can lead to improvements in the treatment of amblyopia patients.

5. Acknowledgements

The authors would like to thank IF Goiano - Campus Ceres for financial for the support, Ótica Cristal, optometry clinic in Goiânia and the Universidade do Estado do Rio Grande do Norte for providing the laboratories for the experiments.

6. References

1. Ribeiro JA, Rodrigues SS, Brasil LM. Occlusion child derived from natural latex for amblyopia treatment. In: *Proceedings of 2013 Pan American Health Care Exchanges (PAHCE 2013)*; 2013 Apr 29-May 4; Medellin, Colombia.

2. Ribeiro JA, Rosa MFF, Rosa LLF, Rocha AF, Rodrigues SS. Oclusão infantil derivada do látex natural para o tratamento de ambliopia. In: *Anais do XXII Congresso Brasileiro em Engenharia Biomédica;* 2010 Nov 21-25; Tiradentes, MG, Brazil.

3. Li J, Thompson B, Ding ZF, Chan LY, Chen X, Yu MB, et al. Does partial occlusion promote normal binocular function? *Investigative Ophthalmology & Visual Science*. 2012;53(11):6818-6827.

4. Vasconcelos GC, Costa MF. Tratamento atual da ambliopia: onde estamos? *Arquivos Brasileiros de Oftomologia.* 2013;76(4):v-vi.

5. Silva MA, Guimarães PCL, Pereira TD, Honório-França AC. Biomateriais e sua biocompatibilidade numa abordagem multidisciplinar na área de saúde, alimentos funcionais e medicina regenerativa. *Revista Eletrônica da Interdisciplinar Univar*. 2012:8(1):87-90.

6. Drewnowska O, Turek B, Carstanjen B, Gajewski Z. Chitosan - a promising biomaterial in veterinary medicine. *Polish Journal of Veterinary Sciences*. 2013;16(4):843-848.

7. Rodrigues SSFR. *Desenvolvimento de um sistema de controle de fluxo esofagiano para tratamento da obesidade*. 1ª ed. São Paulo: Edgard Blücher; 2009. 121 p.

8. Bolognesi LF, Borges FA, Cinman JLF, Silva RG, Santos AG, Herculano RD. Natural Latex Films as Carrier for *Casearia sylvestris* Swartz Extract Associated with Ciprofloxacin. *American Chemical Science Journal*. 2015;5(1):17-25.

9. Pinho ECCM. Uso experimental da biomembrana de látex na reconstrução conjuntival. *Arquivo Brasileiro de Oftalmologia*. 2004;67(1):27-32.

10. Andrade TAM, Iyer A, Das PK, Foss NT, Garcia SB, Coutinho-Netto J, et al. The inflammatory stimulus of a natural latex biomembrane improves healing in mice. *Brazilian Journal of Medical and Biological Research*. 2011;44(10):1036-1047.

11. Ribeiro JA, Rosa SRF, Rosa M. Técnica experimental denominada Van Gogh para confecção de protótipo de lente. In: *Proceedings of the XXIV Brazilian Congress on Biomedical Engineering.* 2014 Oct 13-17; Uberlândia, MG, Brazil; 2014. p. 871-874.

12. Ribeiro JA, Rodrigues SSFR. Modelagem Matemática do Olho Amblíope para Tratamento com Oclusor Derivado de Látex. In: *Anais do VIII Congresso Nacional de Engenharia Mecânica*; 2014 Ago 10-14; Uberlândia MG, Brazil.

13. Cavalcante RAL. *Efeito da adição do líquido da castanhado caju nas propriedades térmicas, ópticas, mecânicas emorfológicas da borracha natural* [Dissertação de mestrado]. Brasília: Universidade de Brasília; 2013.

14. Mrué F. *Substituição do Esôfago Cervical por Prótese Biossintética de látex: estudo experimental em cães* [Dissertação de mestrado]. Ribeirão Preto: Universidade de São Paulo; 1996.

15. Mrué F, Netto JC, Ceneviva R, Lachat JJ, Thomazini JA, Tambelini H. Evaluation of the biocompatibility of a new biomembrane. *Materials Research*. 2004;7(2):277-283.

16. Herculano RD, Guimarães SAC, Belmonte GC, Duarte MAH, Oliveira Júnior ON, Kinoshita A, et al. Metronidazole release using natural rubber latex as matrix. *Materials Research*. 2010;13(1):57-61.

17. Herculano RD, Silva CP, Ereno C, Guimarães SAC, Kinoshita A, Graeffc CFO. Natural rubber latex used as drug delivery system in guided bone regeneration (GBR). *Materials Research*. 2009;12(2):253-256.

18. Mrué F. *Neoformação tecidual induzida por biomembrana de látex natural com polilisina. Aplicabilidade em neoformação esofágica e da parede abdominal. Estudo experimental em cães* [Tese de doutorado]. Ribeirão Preto: Universidade de São Paulo; 2000.

19. Rosa LLF, Alves LAP. *Manual de boas práticas dos estabelecimentos de comércio, adaptação e dispensação de armações, lentes oftálmicas, óculos de proteção solar, lentes de contato e acessórios ópticos.* Rio de Janeiro: Confederação Nacional de Bens, Serviços e Turismo; 2012.

20. Barros NR, Chagas PAM, Borges FA, Gemeinder JLP, Miranda MCR, Garms BC, et al. Diclofenac potassium transdermal patches using natural rubber latex biomembranes as carrier. *Journal of Materials*. 2015;2015:807948.

21. Aielo PB, Borges FA, Romeira KM, Miranda MCR, Arruda LB, L Filho PN, et al. Evaluation of sodium diclofenac release using natural rubber latex as carrier. *Materials Research*. 2014;17(Supl.1):146-152.

22. Agostini DLS. *Caracterização dos constituintes do látex e da borracha natural que estimulam a angiongênese* [Dissertação de mestrado]. São Paulo: Universidade Estadual Paulista Júlio de Mesquita Filho; 2009.

23. Liu C, Shao Y, Jia D. Chemically modified starch reinforced natural rubber composites. *Polymer.* 2008;49(8):2176-2181.

24. Ooi ZX, Ismail H, Bakar AA. Study on the ageing characteristics of oil palm ash reinforced natural rubber composites by introducing a liquid epoxidized natural rubber coating technique. *Polymer Testing*. 2014;37:156-162.