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CHARACTERIZATION ANALYSIS OF NATURAL LATEX MEMBRANES APPLIED IN THE AMBLYOPIC EYE

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Abstract. *The ophthalmic dysfunction called amblyopia consists of low vision in an eye that did not develop properly in childhood. Popularly known as lazy eye, that dysfunction has been one of the most frequent causes of vision loss in children, affecting between 2 to 5% of the population. The products currently used for amblyopia treatment are the conventional occlusors, fixed with tape directly over the eye of the child. They generate aesthetic damage and also enable the child to remove it very often, which brings considerable inefficiency to the treatment. In view of what has been exposed this study presents a simpler approach of eye occlusion based on ophthalmic lens. These lenses derived from a biomaterial occluder that prevents the passage of light from the external environment into the eye. The biomaterial applied is a kind of latex that already presents several applications in tissues with satisfactory results. The lenses made were characterized through physical and chemical methods in order to analyze the characteristics of latex in natura and natural rubber membranes, thermally treated at 40° and 70 °C. Its applicability presents less discomfort when compared with the current products, mainly among children under the age of 6, which are the main victims of amblyopia. The methods used to feature the materials obtained were: X-ray Diffractometry (XRD), Electronic Scanning Microscopy (SEM), Thermogravimetry Analysis (TG) Differential Scanning Calorimetry (DSC), fluorescence and water absorption analysis. Physical and chemical properties analysis of latex membrane used enabled to clarify its application as biocompatible occlusors to be used in children who develop amblyopic eye.*

Keywords: *amblyopia, infant eye occlude, latex membranes.*

1. INTRODUCTION

The amblyopia rates in Brazil are high. Amblyopia is due to functional or organic problems related to vision. According to the World Health Organization (WHO) research indicates that if global outreach initiatives are not taken, by 2020 there will be worldwide 75 million blind people and more than 225 million people with low vision, that is to say, unable to perform large number of daily tasks due to visual impairment (PORTAL BRASIL, 2011).

These diseases may be caused by refractive problems (such as people with high refractive errors in one or both eyes), strabismus (eye motor dysfunction), iatrogenic (generated by incorrect treatments that lead to the appearance of amblyopia in the eye contralateral) and organic. It is seen that the lack of care by professionals and the vast demand for treatment causes the no satisfactory treatment rates increase every day.

According to studies, about 1 in 100 children tend to develop amblyopia and any material or technique that is geared to these cases, which may reach this population, will be able to sensitively lessen the problem. The lack of care and lack of access of the population layers also help to thicken the number of untreated (Sousa, *et al.*, 2012; Reis, *et al.*, 2000).

Amblyopia is the most common visual defects in children, early diagnosis and treatment bring satisfactory results (Salata, *et al.*, 2001; Almeida, 2005). The proposed product will be able to precisely achieve these lower classes, because once developed, it will have more affordable prices as well as greater ease in doing the treatment.

This paper presents in its composition the possibility of generating a new method that does something not yet obtained, in relation to the ocular diseases treatment. The occluding lens may be considered a bracing ('ortho' from Greek) a device applied to any part of the body, alone or covering over a joint, whose function is to stabilize or immobilize, prevent or correct deformity, protect against injury and aid in healing or maximize the function. In this particular case, the occluding lens is placed in the right eye for a few hours or days according to proper treatment made by the ophthalmologist, causing the acceleration of lazy eye view, diagnosed with amblyopia (Ribeiro, *et al.*, 2010; Ribeiro, *et al.*, 2014).

It is noteworthy that the in vivo test was not possible for this study because the proposed innovation. The analyzes presented here serve as a basis for future work.

A biomaterial is a synthetic material or not, used to replace part of a living system or to function in contact with living tissue, aiming to replace, repair, or assist in the function of organs or damaged tissue in a safe, responsible, economically and physiologically acceptable way (Recum and Jacobi 1998; Orefice, *et al*., 2005; Park and Lakes, 2007). The biomaterials are designed to repair and / or reconstruct parts or functions of organs and tissues, so that they may serve as matrix, vehicle, carrier, or enhancer of new tissue formation and / or reconstitution (Ribeiro and Rodrigues, 2014).

A material used in specific applications, develops appropriate tissue response in the host system, is characterized as biocompatible, it does not necessarily have to be completely inert or harmless as previously believed, because the biofunctionality is characterized by performing desired functions, due to its mechanical, chemical, optical, electrical properties, etc. Besides biocompatibility, biomaterials must have biofunctionality, that is, the ability to properly perform the desired function, given its mechanical, physical, and chemical properties. Biocompatibility standard test represent beyond approval criteria and failure to control inserted material in the market, studies parameters of the organism interactions and the material in contact with it, aiming to establish testing standards to describe and grade answers both as host material, eliminating toxicity or setting a tolerance criterion for the risk of material level.

Natural latex from Hevea brasiliensis is a raw material of high quality and durability, has biocompatible physical and chemical characteristics, antigenicity, hypoallergenicity, impermeability, elasticity, softness, flexibility, low cost and resistance, used for making contact lenses (Ribeiro, *et al.*, 2013; Rodrigues, 2009). The choice of latex as material used in lens production called LENCOC®, which is patented and has trademark registration, was based on previous studies research that used latex for medicinal purposes because it is considered a simple material, easy handling and inexpensive. Besides, being induce to neovascularization and tissue repair.

Given this knowledge, this paper presents the production of membrane using latex as a possibility for treating amblyopia shaped as occluding lens and its characterization, using the X-Ray diffraction techniques, scanning electron microscopy (SEM), Thermogravimetry Analysis (TG), differential scanning calorimetry (DSC), Fluorescence and water absorption analysis.

2. MATERIALS AND METHODS

The latex used was purchased in the domestic market. The raw material was purchased from the Southeast suppliers, based on some standard features that were needed such as low sulphur amount and high viscosity. A high sulphur concentration gave the latex, after vulcanization, adhesive and low viscosity characteristics, making the cooking process with high manufacturing time. Therefore, the supplier that met these criteria was decided - bi centrifuged and pre-cured.

From the natural latex was prepared a final compound by addition of chemicals by following the steps proposed in (Mrué, 1996). In order to check the latex membrane (LENCOC®) indispensable features for the lens are elasticity, softness, strength, impermeability and hypoallergenicity. After compound preparation for use in the manufacture of the membrane should follow the steps of filtering and dilution in double distilled water. For this dilution was considered 50% of latex, and 50% distilled water. The whole procedure must referentially be performed at low temperature (below 20 °C) to prevent pre-vulcanization of the liquid, due to the presence of heat.

The development of latex membranes technique was conducted at the Engineering Laboratory and Biomaterial - BioEngLab®, linked to UnB Gama College and characterization tests in the laboratories of Physics and Chemistry at the University of Rio Grande do Norte. This confection process was led in three main steps: preparation of the mould, the membrane preparation and characterization of the product. The characterization process was to observe the behaviour at different temperatures and types of membrane manufacturing.

2.1 Mould Confection

The mould consists of plastics material and coated with an acrylic layer of 3 cm wide, 3 cm long and 1.5 cm in diameter was first made. The mould is designed based on the anatomy and the characteristics of the eye, using as a model the anatomical shape of marble for prototype purposes based on their size, shape and proportion.

For the manufacture of latex blade - it is necessary i) support and ii) mould. Figure 1 illustrates the tecnil bracket for attaching the glass mould used in the lens confections and the mould for generation of latex amine. The making of the membrane is completely individualized; its shape and proportions faithfully follow the characteristics of the patient's eyes, providing greater comfort. Membranes were prepared from the moulds previously washed with soap and water, dried with hot air and sterilized by oven at 70 °C for ten minutes.

Figure 1. Mould the Van Gogh membranes shows A) support, B) glass mould and C) coupling to generate the latex lamina using this technique.

For the deposition technique, was used as template a petri dish measuring 10 cm in diameter according to the literature (Pinho, 2004).

2.2 Membranes manufacture

Van Gogh's technique was based on the immersion technique (Mrué, 1996), which consists on the mould introduction into the final latex compound, perpendicular to the plane, gradually and uniformly, followed by heating in a thermostatic oven. This point represents the polymerization onset, which determines the final manufacture of the product. After this phase, the mould was removed slowly and gradually placed in the greenhouse and subjected to vulcanization at 70 ºC. Then the moulds were removed and membranes were prepared from thick brushstrokes using a flexible rod with cotton tips covered with 1 cm away from the mould with back and forth moves to completely cover the mould. This procedure was repeated three or four times in succession, followed by drying in an oven at 40 °C, with 2 hours of intervals in order to obtain membranes with thicknesses up to 0.2 mm. After this drying period, Van Gogh membranes remained at rest for 24 hours at room temperature to complete the obtaining process. This procedure of obtaining membranes was called Van Gogh, in honour of the Impressionist painter Van Gogh that used a peculiar method of hits with brushes in his works.

The obtaining of membranes by depositing on the petri dish was performed as described in the literature (Mrué, 1996; Mrué, 2000; Pinho, 2004). The transparency of the membrane is favoured in the process by the ambient temperature. The vulcanisation procedure may make the membrane with more yellow appearance, however, for these membranes preparation, which were used on average three to four days for complete drying and curing, because the membrane structure and shape must be very well and evenly preserved, requiring a process more thorough preparation.

Latex membranes obtained by both techniques were developed based on the evolution history with several different protocols to obtain a model that respects the main features: light occlusion, thickness and applicability.

Latex membranes were identified as Van Gogh membranes obtained from three baths (M1) and four baths (M2), both cured at 40 °C, and membranes obtained by deposition technique, cured at 40 °C (M3) and 70 °C (M4).

2.3 Membranes characterization

The latex membranes M1, M2, M3 and M4 obtained were characterized by X-ray diffraction (XRD), scanning electron microscopy (SEM), thermogravimetric analysis (TG), differential scanning calorimetry (DSC), energy dispersive X-ray (EDX) - fluorescence measuring, and water absorption analysis to investigate the latex characteristics of fresh and natural rubber membranes, heat-treated at 40 to 70 °C.

2.3.1 Fluorescence measurements

This technique was used to quantify the components of the membranes and to verify the variation of the pure latex components and cured at 40 and 70 °C.

Fluorescence spectra were performed at room temperature on a fluorescence spectrophotometer SHIMADZU EDX-7000, Energy Dispersive X-Ray Fluorescence Spectrometer.

2.3.2 X-Ray diffractometric (XRD)

The Atomic Spectroscopy is based on the X-ray emission measurements, absorption, scattering, fluorescence and diffraction of electromagnetic radiation. The scattering of X-rays by the crystal ordered environment results in interference between the scattered radiation (constructive or destructive) because the distance between the scattering centers are of the same order of magnitude as the wavelength of the radiation.

X-ray diffractometric (Rigaku Miniflex II RV-200B X-ray diffractometer) of latex membranes was obtained using a Cu-Kα target at 30 kV and 30 mA. The diffraction angle was varied within the range of $5° < 2\theta < 50°$ at a scan rate of $2°\ min^{-1}$.

2.3.3 Scanning Electron Microscopy (SEM)

The scanning electron microscope is generally used for the surface study and subsurface structures of samples with relatively large dimensions, has high depth of focus and can get different reliefs of the sample surface simultaneously in focus. The SEM also produces high-resolution images, which ensures the gain of high magnification of details without losing the sharpness (Agostini, 2009).

Sample preparation is done on metal supports, using conductive adhesives, such as carbon or carbon ribbons colloidal suspensions. The sample coating by a conductive film aims to avoid the accumulation of negative charge in the case of the same is formed by a non-conductive film, as in the case of polymers.

Scanning electron microscopy (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.

2.3.4 Thermogravimetric (TG) and Differential Scanning Calorimetry (DSC)

The TG technique is used to monitor the weight change of a specimen when subjected to a temperature increase at a constant rate. This analysis was done in conjunction with the DSC, which is used to detect physical or chemical changes that are accompanied by heat flow.

When substances are heated, various physical or chemical transformations involving absorption (endothermic process) or evolution of heat (exothermic process) can occur. The DSC technique can be used to investigate thermal properties of materials variety, being used to characterize organic materials, inorganic, organic and polymeric. Among the applications can be mentioned: qualitative and quantitative determination of phase transitions such as the glass transition, melting, crystallization, polymerization kinetics study, decomposition and curing.

Thermogravimetric (TG) analysis, differential thermogravimetric (DTG) analysis, and differential scanning calorimetry (DSC) were performed using a Netzsch Leading Thermal Analysis STA 449 F3 Jupter. All analyses were carried out with a 5 mg sample in alumin pans under a dynamic nitrogen atmosphere between 25 and 800 °C. The experiments were run at a scanning of 10 K/min.

2.3.5 Water absorption analysis

Water absorption measurements were made by immersing the membranes in the latex aqueous solution of 0.9% NaCl and obtained from the mass gain as a function of time. Samples equivalent to an area of 1 cm^2 were hydrated and maintained in 25 ml solution for 24 hours at room temperature throughout the experiment.

3. RESULTS

This section will present the results of characterization of latex in natura, and natural rubber occluding lens membranes made by Van Gogh technique with three baths and four showers heat treated at 40 °C and the deposition technique in the petri dish 40 °C and 70 °C as described in the literature (Mrué, 1996), using the techniques of x-ray diffraction (XRD), scanning electron microscopy (SEM), TG, DSC, fluorescence and absorption.

3.1 X-ray diffraction

On the X-ray diffraction technique, the membranes used were the Van Gogh three and four baths at 40 °C and the petri dish membrane 40 to 70 °C, as shown in Figure 2 in order to verify the presence of crystalline compounds, such as the presence of metals or structures with a certain organization.

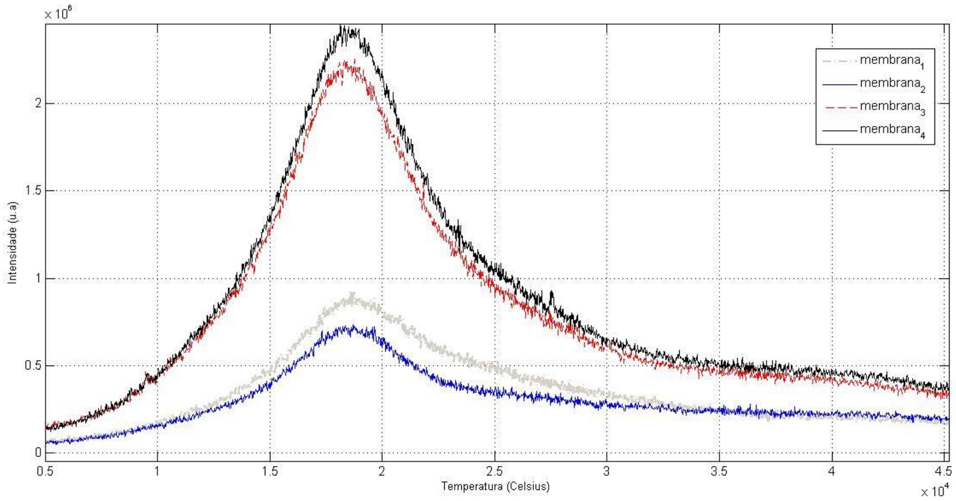
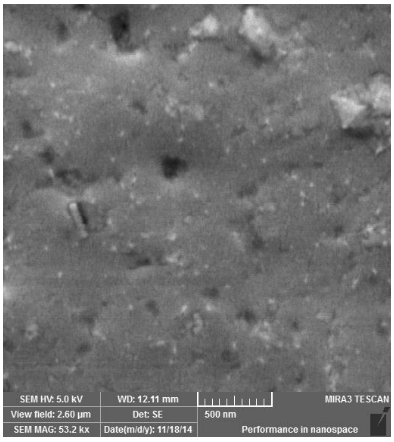


Figure 2. X-ray diffraction of the membranes M1, M2, M3, M4 3.

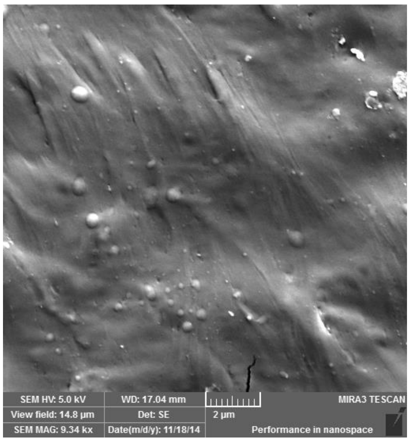
In the diffractogram of the membranes M1, M2, M3 and M4 we have the absence of diffracted peaks that characterize a crystalline phase, there was only a broad peak centred at 2θ scattering = 19 °C, which is consistent with that previously reported in literature (Agostini, 2009), assigned to x-ray diffraction in the amorphous regions of the samples. The membranes M3 and M4 had an X-ray diffraction profile with some crystallinity, as compared to membranes M1 and M2, although it has been prepared by deposition technique at different vulcanising temperatures.

3.2 Scanning Electron Microscopy (SEM)

Figure 3 shows the micrograph with magnification of 8000 times above the natural rubber membranes thermally treated at 40 °C and 70 °C, and (a) M1, (b) M2, (c) M3 and (d) the M4. The M1 shows roughness, small porosity can be observed along the cut that penetrates the pores across the membrane allowing water absorption. Notice that the micrograph M2 has rubber particles, coagulated quickly with an extra bath leaving the particles dense, and presence of a porous distribution both along the cutting surface and in the pores measuring less than 1 micron. On the M3 shows the presence of more organized and dense rubber particles. The M4 presents be translucent and is not recommended for closure, in which the presence of high temperatures during vulcanization synthesize the pores making them larger, there was a higher uptake in shorter time.



a



b

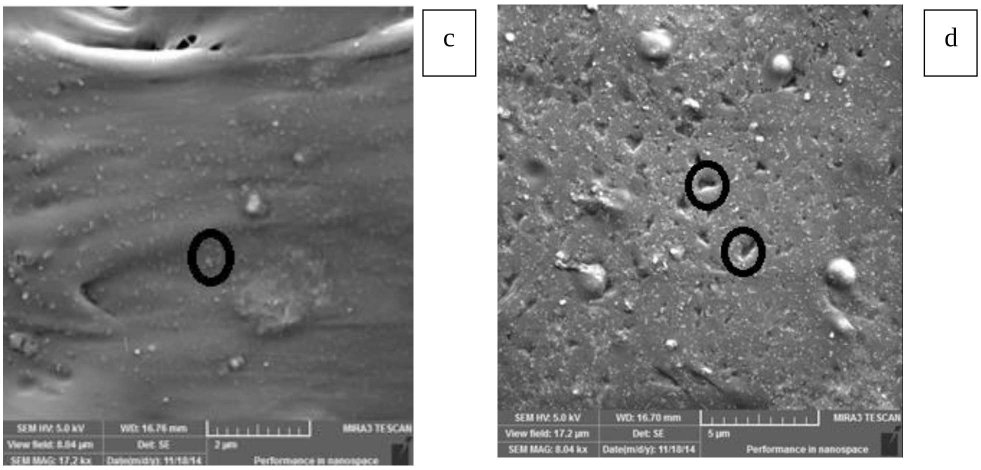


Figure 3. Micrograph M1 membranes (a), M2 (b), M3 (c) and M4 (d).

3.3. Thermogravimetric (TG) and differential scanning calorimetry analyst (DSC)

Figure 4 shows the DSC and TG curve of the latex produced membranes at 40 °C and 70 °C. We showed complete degradation at a temperature about 300 °C, temperature which is consistent with the onset of thermal degradation of the membranes.

The membranes M1, M2, M3 and M4 had a low weight loss. This weight change was very smooth, as can be seen by the low variation in the DTG in that range. This should have happened due to the thermal treatment in an oven latex, wherein the low thermal stability of components can be evaporated or more strongly bonded to the rubber particles (Agostini, 2009). The fact that the latex used to be bi-centrifuged also contributes to this behaviour. It can be noticed that the IGT higher weight loss began at 290 ° C after the start pyrolysis of the polymer. As given by (Agostini, 2009), a variation in IGT to about 420 °C was observed, indicating degradation of the polymeric chains thermally more stable.

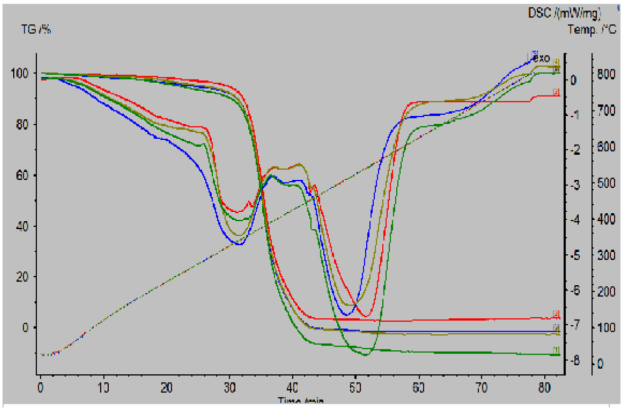


Figure 4. Curves M1 TG and DSG membranes (Red), M2 (green) and M3 (blue) and M4 (yellow).

3.4 Fluorescence

The technique of energy dispersive X-ray (EDX) was performed in order to identify the chemical elements of the membranes after the preparation process and vulcanization of the composite. Table 1 shows the values of the elements found in samples M1, M2, M3 and M4.

Table 1. Membranes chemical elements EDX

	Sulphur (S)	Potassium (K)	Phosphorus (P)	Zinc (Zn)	Others
Membrane 1 (M1)	79%	2%	17%	1%	1%
Membrane 2 (M2)	70%	26%	2%	1%	1%
Membrane 3 (M3)	70%	21%	1,5%	6%	1,5%
Membrane 4 (M4)	58%	20%	1,5%	19%	1,5%

On Table 1, we see the presence of elements S (Sulfur), K (potassium), P (Phosphorus), Zn (zinc), Cu (copper) in all membranes, and the more intense values are assigned to S, K. other amounts of chemicals are small in all the membranes M1, M2, M3 and M4.

M1 membrane presented higher concentrations of sulfur than other membranes because of its technical and Van Gogh the least amount of latex in its curing process. M4 had a lower amount of sulfur, which underwent vulcanization at 70 ° C and it increased the zinc in relation to M1 19%.

3.5 Absorption

Table 2 shows the weights of the membranes observed at different time intervals. This analysis showed that the M1 membrane gained greater mass a larger time interval while M4 membrane of 70 °C had a greater stability in a shorter time interval. The increase of membranes' water absorption associated consequently have lower crystallinity than the crystalline regions constitute barriers to penetrate water. Among the temperatures of 40 °C, the Van Gogh technique with 4 baths had a lower water absorption.

Table 2. Membranes' water absorption with respect to time

Sample	Time (minutes)	Absorbed Mass
Membrane 1	375	5,4%
Membrane 2	105	4,4%
Membrane 3	105	9,0 %
Membrane 4	30	1,5%

4. CONCLUSIONS

Initial studies in this paper demonstrated the feasibility of using membrane preparations of Van Gogh technique in relation deposition technique. The initial focus of this study was to characterize the latex membrane for use as an occluder for treating amblyopia. The results show strong evidence that the membranes prepared by the technique of Van Gogh 4 baths at 40 °C appears as a diffuse light passing the membrane, thus causing the occlusion.

The importance of biosafety has been exercising in practice for bioengineering linked to medicine stimulates new research proving the efficacy of the methods, since many treatments for the approached disease are considerable unacceptable.

The ability to generate other results from these in future work will be a line of action that the authors intend to explore and other works that are under development, including statistical analysis and exploitation of data.

The relevance of the study performed is clear, since many treatments for amblyopia are considered unacceptable. Therefore, the development of natural latex membranes can lead to improvements in the treatment sighted patients.

5. REFERENCES

Almeida, L.O.C. 2005. "A cost analysis of therapy for amblyopia for an outpatient at a university hospital". *Arquivo Brasileiro de Oftalmologia*, 68(4): 475-480.

Agostini, D.L.D.S. 2009. *Caracterização dos constituintes do látex e da borracha natural que estimulam a angiongênese*. Dissertação de Mestrado, Universidade Estadual Paulista Júlio de Mesquita Filho (UNESP), Bauru – Presidente Prudente.

Mrué, F. 1996. *Substituição do Esôfago Cervical por Prótese Biossintética de látex: estudo experimental em cães*. Dissertação de mestrado, Universidade de São Paulo, Faculdade de Medicina da Ribeirão Preto, Ribeirão Preto.

Mrué, F. 2000. *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, Universidade de São Paulo, Faculdade de Medicina de Ribeirão Preto, Ribeirão Preto.

Orefice, R.L., Pereira, M.M., Mansur, H.S. 2005. *Biomateriais: Fundamentos E Aplicações*. Ed. Cultura Médica, Rio de Janeiro, 1° edição.

Park, J.B. and Lakes, R.S. 2007. *Biomaterials: an introduction*. Ed. Springer Science corp., New York, 3ª edition.

Pinho, E.C.C.M. 2004. "Experimental use of latex biomembrane in conjunctival reconstruction". In: *Arquivo Brasileiro de Oftalmologia*, v.67, n.1, 27-32.

Recum, A.V. and Jacobi, J.E. 1998. *Handbook of biomaterials evaluation: scientific, technical, and clinical testing of implant materials*. Ed. Edwards Brothers, 2ª edition.

Reis, P.P.L., Kawakami, W.O., Mourão, D.A.C. 2000. "The use of Photoscreening in the study of the amblyopigenics factors in children". *Arquivo Brasileiro de Oftalmologia,* 63(1): 45-48.

Ribeiro, J.A., Rosa, M.F.F., Rosa, L.L.F., Rocha, A.F., Rodrigues, S.S. 2010. “Occlusion Child Natural Latex Derivative for Amblyopia Treatment”. In: *XXII Brazilian Congress on Biomedical Engineering, Proceedings of the XXII Brazilian Congress on Biomedical Engineering,* Tiradentes, MG, Brazil.

Ribeiro, J.A., Rodrigues, S.S., & Brasil, L.M. 2013. “Occlusion child derived from natural latex for amblyopia treatment”. In: *Pan American Health Care Exchanges (PAHCE) Conference, workshops, and exhibits Cooperation/Linkages*, Medellin, Colombia. IEEE, 2013. v. 1. p. 329-329.

Ribeiro, J.A**.**, Rosa, S.R.F., Rosa, M. 2014. “Experimental technique designated Van Gogh for making lens prototype”. XXIV *Brazilian Congress on Biomedical Engineering, Proceedings of the XXIV Brazilian Congress on Biomedical Engineering*, Uberlândia, MG, Brazil, v. 1, p. 871-874.

Ribeiro, J.A. and Rodrigues, S.S.F.R., 2014. “Modelagem Matemática do Olho Amblíope para Tratamento com Oclusor Derivado de Látex”. *VIII Congresso Nacional de Engenharia Mecânica*. Uberlândia, MG, Brazil.

Rodrigues S.S.F.R. *Desenvolvimento de um sistema de controle de fluxo esofagiano para tratamento da obesidade*. São Paulo: Edgard Blücher Ltda, v. 1. 121 p., 1° edição.

Salata, A.C.F., Villaça, V.T.N., Roma, R.L., Norato, D.Y.J. E Carvalho, K.M.M. 2001. “Occlusion therapy in amblyopia: factors that influence the outcome”. *Arquivo Brasileiro de Oftalmologia,* 64:123-6.

Sousa, R.L.S., Funayama, B.S., Catâneo, L., Pandovani, C.R., Schellini, S.A. 2012. “Comparison between visual acuity and photoscreening used like visual screening methods for scholar aged children”. *Journal of Ophthalmology*; 71 (6): 358-63.

PORTAL BRASIL, 2011. “Agência Brasil”. 19 may. 2015. <http://memoria.ebc.com.br/agenciabrasil/noticia/2011-10-10/diagnostico-precoce-diminui-riscos-de-perda-de-visao-em-criancas>.

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6. RESPONSIBILITY NOTICE

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