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UTILIZATION OF NANOPARTICLES OF POLY (LACTIC-CO-GLYCOLIC ACID) (PLGA) IN THE NEUTRALIZATION OF SARS-CoV-2: REVIEW

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Abstract: SARS-CoV-2 has gained prominence as it has become a threat to human health and the ecosystems worldwide. In the Materials Engineering field, researches have observed the ability of nanoparticles to neutralize the infection potential of viral entry into host cells. Hence, we highlight the poly nanoparticles (lactic-co-glycolic acid) (PLGA) due to their high antiviral action, biodegradability, biocompatibility, and low toxicity, which is widely used in the development of nanomedicine and also as a new alternative to combat viral diseases in the current context of SARS-CoV-2. In this scenario, this research consists in a literature review that aimed to identify the antiviral activity of poly nanosponges (lactic-co-glycolic acid) (PLGA) in the neutralization of SARS-CoV-2. The results indicate a virucidal effect of poly nanosponges (lactic-co-alycolic acid) (PLGA) against SARS-CoV-2 by around 90%, since poly nanosponges (lactic-co-glycolic acid) (PLGA) attach to the spike of SARS-CoV-2, preventing viral entry in the cell membrane.
Keywords: SARS-CoV-2, Poly Nanosponges (lactic-co-glycolic acid) (PLGA), Nanoparticles.

INTRODUCTION

SARS-CoV-2 (acute severe respiratory coronavirus syndrome 2) is the virus responsible for coronavirus disease of the year 2019 (COVID-19). This disease was first identified in Wuhan (Hubei, China) in December 2019, and it was declared a pandemic by the World Health Organization (WHO) in March 2020 (World Health Organization, 2020; Centers for Disease Control and Prevention, 2020).

SARS-CoV-2 is a virus of the coronavirus family that causes the respiratory disease called COVID-19. The SARS-CoV-2 belongs to the order *Nidovirales*, family *Coronaviridae*, subfamily *Orthochoronaviriae* and gender *Betacoronavirus* (International Committee on Taxonomy of Viruses, 2019). The transmission of diseases between humans occurs mainly through respiratory droplets (Centers for Disease Control and Prevention, 2020).

The coronavirus family (*Coronaviridae*) is in the range of 65 to 125 nanometers (nm) of diameter, having only one single RNA tape (size range 26 to 32 kbs long) as nucleic material (Shereen et al., 2020). Hence, it is divided into 4 genera: *Alphacoronavirus* (α-CoVs), *Betacoronavirus* (β-CoVs), *Gammacoronavirus* (γ-CoVs) e *Deltacoronavirus* (δ-CoVs) (Wang et al., 2018).

SARS-CoV-2 consists of four types of structured proteins, which are spike glycoprotein (S), membrane protein (M), envelope proteins (E) and nucleocapsid (N) (Figure 1) (Florindo et al., 2020). Among these, the spike glycoprotein (S) plays a key role in binding the virus to the host cell and, later, in the cell entry (Patil, 2020).

With the mechanism of coronavirus infection in host cells, the most affected organ is the lungs. The angiotensin-converter enzyme 2 (ACE-2) is found in large numbers in alveolar cells type II of the lungs (Hogan, 2011). Once in the respiratory tract, SARS-CoV-2 binds to lung cells by endocytosis via angiotensin-converter enzyme 2 (ACE-2) (Cui; Li; Shi, 2019).

The alveolos are small shell-shaped cavities located in the pulmonary structure, where gas exchange of the respiratory process takes place. Type II cells operates at the formation of the alveolos. Thus, the airways are the potential gateway to virus infection (Letko; Marzi; Munster, 2020). Figure 2 shows the process of coronavirus infection and the replication cycle.

Spike glycoprotein interfaced with the angiotensin-converter enzyme 2 (ACE2) to

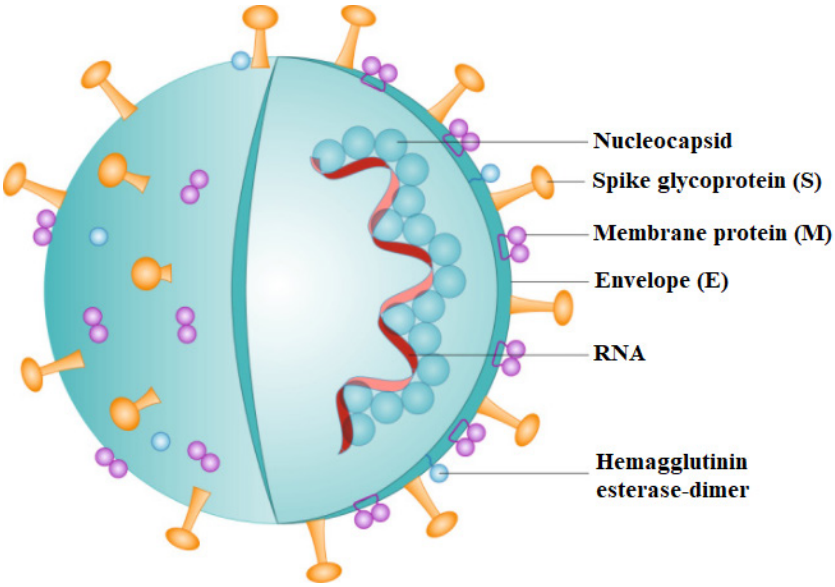

Figure 1: Schematic representation of SARS-CoV-2 structure. This is an enveloped, positive-sense RNA virus with four main structural proteins, including spike (S) and membrane (M) glycoproteins, as well as envelope (E) and nucleocapsid (N) proteins (Adapted from Florindo *et al.*, 2020).

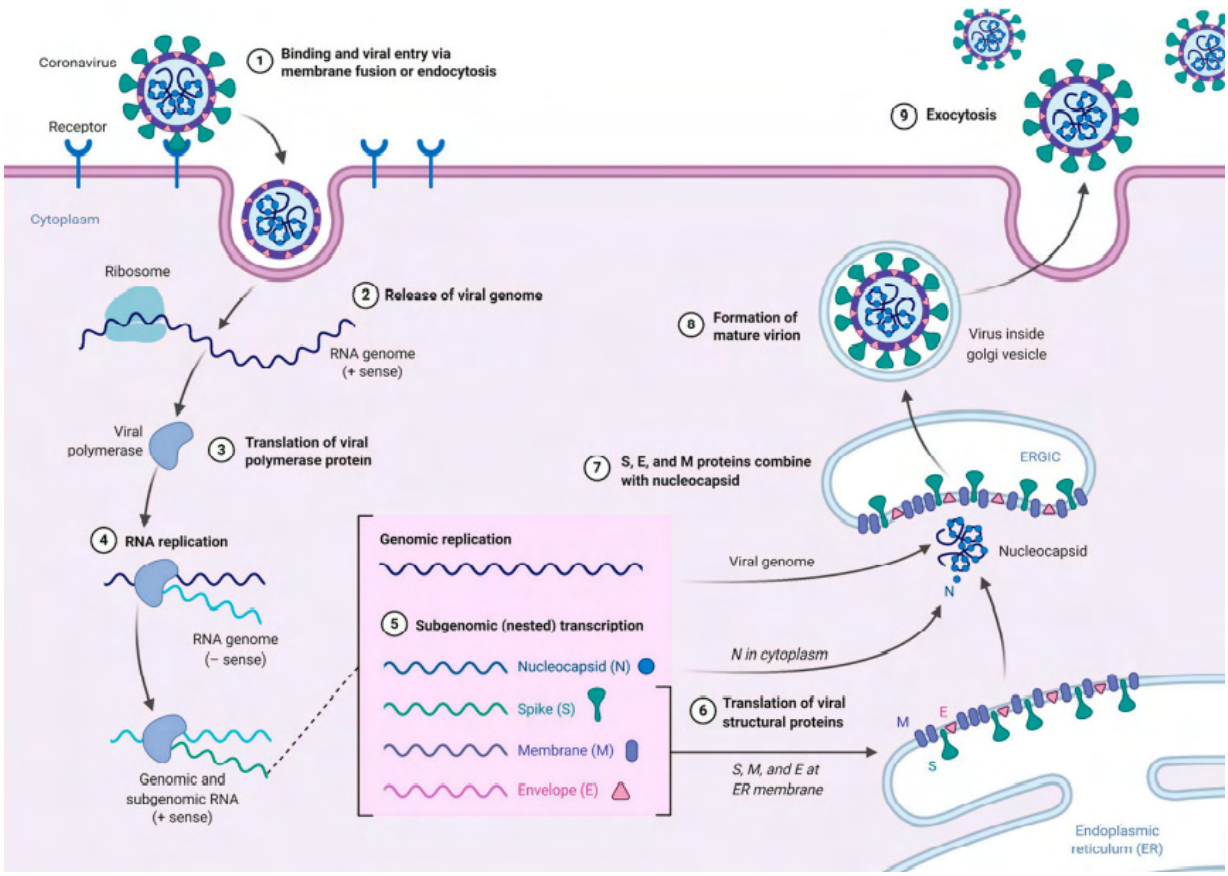

Figure 2: Coronavirus infection mechanism and replication cycle (Bilal *et al.*, 2020).

join with host cells. The envelope protein contributes in the setting and formation of new viridions in the endoplasmic reticulum. The membrane protein is the main structural protein. The nucleocapsid protein binds to viral RNA and it is included in the processing and stabilization of RNA (Astuti; Ysrafil, 2020; Nieto-Torres et al., 2011).

The ability of microorganisms to trigger disease in humans and in the environment has become a global threat, mostly due to their speed of propagation. Thus, there is a need to improve and develop biocompatible control alternatives that have antiviral agents and reduce, for example, SARS-CoV-2 infection, especially in critical areas such as health units and public places. In this context, the use of nanotechnology and nanomaterials has become a trend, being a reality to this issue (Vazquez-Munoz; Lopez-Ribot, 2020).

Nanoparticles consist of particles or clusters of different sizes, which have a significant fraction of particles with one or more dimensions in the range between 1 and 100 nanometers (nm) (Schulz, 2013). They are applied, for example, in the fields of biology, chemistry, biomedicine and materials science (Rotello, 2004). Thus, nanoparticles can be presented in various forms such as silver nanoparticles (AgNPs), gold nanoparticles (AuNPs), polymeric nanoparticles, quantum dots (QDs), carbon points (CDots), graphene oxide (GO), silicon materials, dendrines, liposomes and others (Gurunathan et al., 2020).

Among the main advantages in the use of nanoparticles, it is important to mention the controlled and/or prolonged release of the substance incorporated into them. There is also a decrease of adverse effects associated with the substance, the protection of neutralization compounds before reaching the means of action and the growth of intracellular penetration (Santos; Fialho, 2007). Currently, there is a satisfactory evolution in the field of science and technology regarding the use of nanoparticles. Thus, it is essential to highlight the variety and importance of selective nanoparticles that can be used as antiviral agents such as polymeric nanoparticles.

Poli (lactic-coglycolic acid) (PLGA) is a synthetic, biodegradable, biocompatible copolymer formed by means of condensation reactions between lactic acid and glycolic acid monomers, in various proportions (Figure 3). In addition, this copolymer has been approved by the Food and Drug Administration (FDA) and European Medicines Agency (EMA). It has been widely used in biomedical areas such as drug and implant release systems (Mir; Ahmed; Rehman, 2017), since it has properties such as its faster complete degradation by hydrolysis (Motta; Duek, 2006), regular chain geometry, mechanical resistance, sustained release and appreciable safety profile, providing low toxicity and biocompatibility (Erbetta et al., 2012; Kolate et al., 2015).

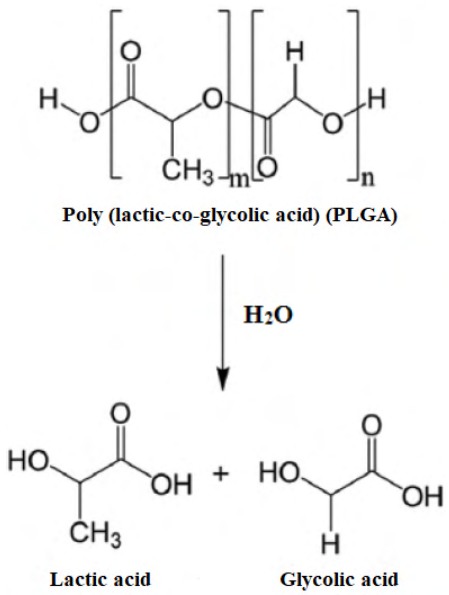

Figure 3: Chemical structure of PLGA and its degradation products. In the representation m and n refer to the monomeric units of lactate and glycolate present in the copolymer (Adapted from Dumitru *et al*., 2015).

A variation from many days to months is common for the PLGA degradation, depending on the composition and molecular weight of the copolymer, the molar ratio of the monomers, the degree of crystallinity, the glass transition temperature of the polymer and the size of the particles (Erbetta et al., 2012; Yuan; Gu, 2015; Sharma et al., 2016). Normally, PLGA has a vitreous transition temperature (Tg) ranging from 40 to 60°C. In this condition, it is maintained in a thermoplastic amorphous polymeric matrix below the glass transition temperature (Champion; Katare; Mitragotri, 2007).

In this perspective, the biodegradable poly polymeric nanoparticles (lactic-coglycolic acid) (PLGA) have become a fascinating strategy to develop the applicability of antimicrobial activities. They became alternatives to face viral diseases, with emphasis on nanosponges.

Nanosponges consist of small sponges with porly structures. They are classified as nanoparticles due to its size, similar of a virus, with an average diameter below 1 ¼m. Nanosponges have a three-dimensional network, due to their reduced size and porly nature, these pore particles can bind to unsolvable drugs within the matrix and improve their bioavailability by modifying the pharmacokinetic parameters of the drug molecules (Bezawada et al., 2014; Ajinkya; Prakash; Vishal, 2015).

The choice of poly nanoparticles (lactic-coglycolic acid) (PLGA) in this study is justified by their excellent biodegradability, biocompatibility, mechanical resistance, low toxicity, stability under physiological conditions and facility in the controlled release of drugs, being widely used in the development of nanodrugs (Kamaly et al., 2016). In addition to that, they present antibacterial, antiviral and antifungal actions (Jahagirdar et al., 2019), which has become a new alternative to combat viral diseases in the current context of the COVID-19 pandemic. From these researches, there is a new path to inactivation of these viruses and, consequently, the possibility of a cure for the coronavirus. In this scenario, nanotechnology has been developing nanoparticles in order to inactivate SARS-CoV-2, and the results are promising. The choice for these nanoparticles was due to the increasing use of microparticles in the current context, besides the innovative, promising, challenging character, and importance in the area of biomaterials in the fight of COVID-19. Therefore, the general objective of the research was to identify the antiviral activity of poly nanosponges (lactic-coglycolic acid) (PLGA).

METHODOLOGY

The present study was carried out through a bibliographic survey, conducted between September 2020 and September 2021, using the literature available in several databases such as: ScienceDirect, SciELO (Scientific Electronic Library Online), Research Gate and Google Scholar, considering Portuguese and English language materials.

The descriptors used in the Portuguese were: Microorganisms; virus, SARS-CoV-2, coronavirus, COVID-19, Mechanism of Action of SARS-CoV-2, nanotechnology, nanomaterials, nanoparticles, polymeric nanoparticles, biodegradable polymeric nanoparticles, Poly(lactic-co-glycolic acid) (PLGA).

The criteria of inclusion used for the search consisted in scientific articles published in the last 17 years, including national and international manuscripts with the objective of describing updated data on the factors that contribute to the bactericidal and virucide potential of PLGA nanosponges. Exclusion criteria apply to articles that did not address the theme above, or literature

published before 2016.

In this study, articles originated from research and review were selected, as well as books and magazines between 2004 and 2021. The most recent words were prioritized so that the review was as updated as possible. The articles obtained were evaluated according to their relevance, that is, the ones which presented topics directly related to the subject. The data was described in the form of tables, graphs and figures.

RESULTS AND DISCUSSION

In this section, three experimental studies in English on the antiviral activity of poly nanosponges (lactic-coglycolic acid) (PLGA) in the inactivation of SARS-CoV-2 were used. However, no articles in Portuguese or Spanish were used based on this theme. Regarding the sequential analysis of these articles, an average of 35 scientific articles and 4 reference sites were used. After reading the selected references, the information was extracted and organized in the form of a bibliographic review.

ANTIVIRAL ACTIVITY OF POLYNANOSPONGES (LACTIC-COGLYCOLIC ACID) (PLGA)

In the study by Tan et al. (2021) a system for the release of biomimetic drugs based on nanocarriers against the SARS-CoV-2 virus was developed through anti-inflammatory and antiviral treatment simultaneously. These are antiviral agents, hence, the biomimetic lopinavir (LPV) present in poly nanoparticles (lactic-coglycolic acid) (PLGA-LPV) biomimetics of macrophages, membranes from human macrophage cell line (THP-1 cells) were fused into poly nanocores (lactic-co-glycolic acid) (PLGA NPs) by sonication.

These biomimetic substances studies involve the structure, formation or function of compounds and materials made biologically (such as enzymes or seda) and biological methods, such as photosynthesis or protein synthesis, mainly with the objective of synthesizing similar products by artificial procedures that copy natural ones (Bhushan, 2009). Macrophages consist of large leukocytes and widely versible that act intrinsically, with the purpose of performing phagocytosis and being the cellular defenses of the body in inflammatory processes (Watanabe et al., 2019; Hamidzadeh et al., 2017).

Moreover, the morphological analysis of biomimenolic poly (lactic-coglycolic acid) nanoparticles were corred with uranila acetate and visualized by transmission electron microscopy (MET) in the study by Tan et al. (2021). The image obtained found that the biomimetic PLGA nanoparticles of macrophages are spherical in the surrounding form with a monolaser membrane, which presented a typical nucleus-shell shape. After the membrane coating, it was observed that the sizes of the nanoparticles grew from 85.8±4.4 nm to 102.2±4.0 nm, and the zeta potential of surface of the nanoparticles decreased from 42.4±1.7 mV to 12.4±1.0 mV.

In addition, western blot (method based on the separation of proteins by molecular weight by means of an electrophoresis) demonstrated that the main cytokine receptors related to cytokine release syndrome (Tan et al., 2021), including interleukins (which are protein substances basically generated by leukocytes that trigger inflammatory processes in the body) (Germolec et al., 2018) of IL-6 (IL-6R) and IL-1 (IL-1) and IL-1 (IL-1R), and the crucial ACE II receptor of SARS-CoV-2, were expressed in biomimetic PLGA NPs of macrophages, which implies the potential application of biomimetic PLGA NPs of macrophages in the sars-cov-2 neutralization. It was observed that the absorption peaks of PLGA-LPV and PLGA-LPV biomimetic macromimetics were consistent with LPV,

demonstrating that the LPV was successfully encapsulated in the biomimetic PLGA and PLGA NPs of macrophages. The LPV release profiles of biomimetic PLGA and PLGA NPs of macrophages were recorded using a UV-Vis spectrophotometer at the wavelength of 267 nm (Tan et al., 2021).

The LPV release rate of biomimetic PLGA of macrophages was lower than that of membranes not coated with PLGA NPs. Meanwhile, the cumulative release of biomimetic PLGA LPV from macrophages was 71.5% less than 95.3% of PLGA NPs in 24 h, proposing a longer release of biomimetic PLGA from macrophages because of coated membranes. It is importante to mention that PLGA-LPV biomimetic macrophage presented a meaningful stability of nanoparticle size over the course of 72 hours when suspended in water, 1 × PBS and 50% serum, respectively (Tan et al, 2021).

It was possible to analyze in the study by Tan et al. [31] the use of mouse hepatitis virus (MHV) also known as mouse coronavirus (MCoV) as a surrogate virus for SARS-CoV-2. This experiment aimed to verify the antiviral activity of lopinavir contained in poly polymeric nanoparticles (lactic-coglycolic acid) macrophage sars, since it is equivalent to SARS-CoV-2. Furthermore, MHV was initialized as an ideal substitute to verify the neutralization of SARS-CoV-2 by UV-C treatment in the study by Pendyala et al. (2020) Therefore, MHV was used in the study by Tan et al. (2021) as a substitute virus for SARS-CoV-2 to verify the antiviral activity of PLGA-LPV biomimetics of macrophages.

In the study by Tan et al. (2021), macrophages derived from rodents (RAW264.7 cells) were selected as the source of biomimetic PLGA-LPV membranes of macrophages to match the mouse model virus. Similarly, 4 mg/mL of rodent macrophage from macrophage biomimetic PLGA showed a cytokine removal yield of 98% for mouse IL-6 and 63.8% for mouse IL-1², which means that macrophage biomimetic PLGA also had an ability to neutralize mouse pro-inflammatory cytokines.

Later, in the study by Tan et al. (2021), MHV was first incubated with PLGA-LPV biomimetics of macrophages at different doses individually for 1 hour, and then the mixtures were added to l929 fibroblast culture plates for 24 hours of incubation.

Considering that LPV is a lipophilic drug (which has affinity with lipids), free LPV is unstable in aqueous solution. Then, water-soluble PLGA-LPV NPs were used in this study as a control group. Thus, both PLGA-LPV and PLGA-LPV biomimetic macromimetics of macrophages were able to avoid viral multiplication depending on the dose. The viral charge in the group treated with biomimetic PLGA-LPV macrophages was rapidly lower than the PLGA-LPV Group of NPs. Tan et al. (2021) also observed that the biomimetic PLGA-LPV of macrophages could present a better affinity to the virus due to the macrophage membranes.

In conclusion, Tan et al. (2021) evidenced that lopinavir (LPV), as a model of antiviral drug, was brought into polymeric nanoparticles (PLGA-LPV NPs) first. Subsequently, macrophage membranes were coated by poly polymeric nanoparticles (lactic-co-glycolic acid) (PLGA) aggregated with lopinavir (LPV) to constitute biomimetic macrophage nanocarriers. It was observed in the study by Tan et al. (2021) that the poly polymeric nanoparticle (lactic acid-coglycolic acid) containing lopinavir was able to neutralize several pro-inflammatory cytokines and effectively suppress the activation of macrophages and neutrophils. In addition, the production of serum-induced neutrophil extracellular from patients with SARS-CoV-2 could also be decreased by the

poly polymeric nanoparticle (lactic-coglycolic acid) which contained lopinavir. In a murine model of coronavirus infection, biomimetic PLGA-LPV of macrophages demonstrated significant capacity directed to the sites of inflammation, in addition to a superior therapeutic efficacy in relieving inflammation and reducing viral loads in tissues.

In the study of Pourhajibagher et al. (2021), the ability of antimicrobial photodynamic therapy was verified and evaluated as an approach to adjuvant therapy and treatment using curcumin-poly nanoparticles (lactic-coglycolic acid) to neutralize the SARS-CoV-2 epidemic in plasma. Therefore, it was preferable to verify the changes in levels of coagulation aspects, the required quality of plasma treated with antimicrobial photodynamic therapy and the total of protein plasma and anti-A and/or anti-B antibody titrations in the patient's plasma before and after treatment with antimicrobial photodynamic therapy.

The stability of poly curcumin nanoparticles (lactic-coglycolic acid) was determined by particle size around 30 days at 4 °C and 37 °C in the study by Pourhajibagher et al. (2021). There was not a significant variation between days 0 and day 30 ($p > 0.05$). No significant change was achieved in the polydispersity index ($p > 0.05$) and the zeta potential ($p > 0.05$) for 30 days at 4 °C and 37 °C. Thus, no aggregation was verified. With the results obtained in laboratory tests, the study by Pourhajibagher et al. (2021) evidenced the presence of SARS-CoV-2 (84.3%) with gene amplification in the synthesized cDNA model.

To achieve the results obtained, Pourhajibagher et al. (2021) determined the subcytotoxic effect (maximum dosage level before it became toxic to the body) of antimicrobial photodynamic therapy for the treatment of plasma with the presence of SARS-CoV-2, the cytotoxicity of Vero cells of various concentrations of curcumin-poly nanoparticles (lactic-coglycolic acid), different energy doses of blue laser and antimicrobial photodynamic therapy with several doses of blue laser energy (104.5, 313.7 and 522.8 J/cm²) in combination. The concentrations of curcumin-poly nanoparticles (lactic-co-glycolic acid) (3, 5, 7, 10 and 15%) were studied. Different concentrations of curcumin-poly nanoparticles (lactic-coglycolic acid) (3, 5, 7 and 10%) isolated did not have an effect on viable cells (1.5, 4.0, 5.8, 6.8 and 23.4% reduction, respectively).

A statistically significant cytotoxic effect (23.4%) was caused by the increase in the concentration of curcumin-poly nanoparticles (lactic-coglycolic acid) to 15% compared to the untreated control group ($P < 0.05$. That was not the desired effect in relation to curcumin-poly nanoparticles (lactic-co-glycolic acid). However, laser energy doses of 104.5, 313.7 and 522.8 J/cm² caused around 2.8, 4.7 and 10.9% respectively to decrease in the viable cell when applied alone. Vero cells were infected with plasma treated by antimicrobial photodynamic therapy at the highest non-cytotoxic concentration of curcumin-poly nanoparticles (lactic-coglycolic acid) (10% by weight) and the highest energy density of blue laser light (522.8 J/cm²) (Pourhajibagher et al., 2021).

Based on laboratory results obtained by Pourhajibagher et al. (2021), it was observed that there was a significant decrease in cells in Vero cells treated with plasma presenting SARS-CoV-2 using 10% by weight. In addition, the treated plasma containing SARS-CoV-2 with antimicrobial photodynamic therapy (10% by weight of curcumin-PLGA nanoparticles plus blue laser light at 522.8 J /cm²) showed no cytotoxic effect on the Vero cell line. To sum up, Pourhajibagher et al. (2021) found that antimicrobial photodynamic therapy (10% by weight of curcumin-PLGA nanoparticles in combination with blue laser light at 522.8

J/cm²) had an anticoronavirus effect in vitro without the cytotoxic effects in Vero cell culture by a treated plasma containing SARS-CoV-2.

Moreover, in the study by Zhang et al. (2020) microscopic poly cellular sponges (lactic-coglycolic acid) (PLGA) were developed, showing the capacity of neutralizing the activity of the SARS-CoV-2 virus. These nanosponges act as biological imitations or baits to which the virus would bind instead of the host cell. They are coated with cell membranes extracted from macrophages and pulmonary epithelial cells with receptors of the angiotensin-converter enzyme 2 (ACE2) and differentiation cluster 147 (CD147), a transmembrane glycoprotein receptor also found in a soluble plasma form) (Egawa et al., 2006), which binds to the Spike protein found in SARS-CoV-2, responsible for the entry and contagion of the virus.

Additionally, the cell membranes of these microscopic sponge pieces are coated in poly polymeric nanoparticle nuclei (lactic-coglycolic acid) (PLGA). In this work, two types of nanosponges were produced: one with membrane produced with human lung epithelial cells type II, and the other with membrane formed with human macrophages, respectively (Figure 4) (Zhang et al., 2020).

The results showed that, after incubation with nanosponges (both lung cells and immune system), SARS-CoV-2 virus activity in cell culture was reduced by approximately 90, depeding on the dose. This data verifies a high antiviral efficacy in vitro. That is, the use of nanosponges led to the blocking of the virus entry into the cell, which can considerably decrease the chance of someone developing the disease. These are promising data for the inhibition of viral activity. Another significant aspect of this strategy is that it is not related to the virus's ability to mutate (Zhang et al., 2020).

Zhang et al. (2020) demonstrated that the superficial polymeric structure of PLGA is coupled to the coronavirus spicule, also called Spike. This coupling prevents the spicule from interacting with the cell membrane, preventing the virus from reaching the cell and functioning as a kind of barrier. This may be a justification for the use of PLGA

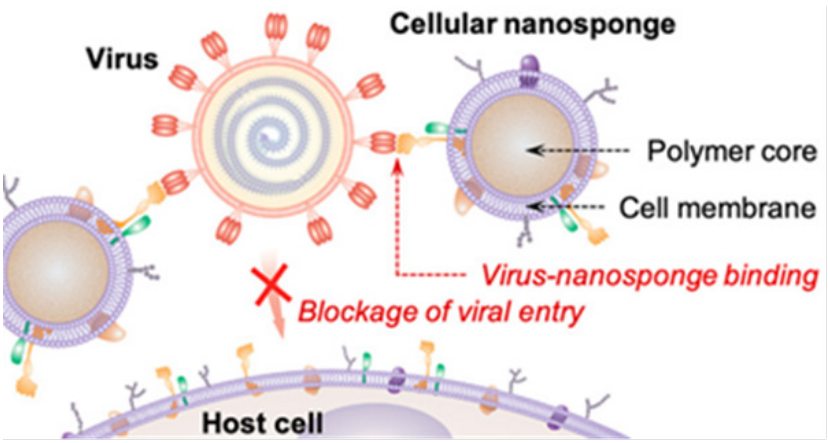

Figure 4: Schematic mechanism of cellular nanosponges that inhibit SARS-CoV-2 infection. The nanosponges were constructed involved polymeric nanoparticle nuclei with natural cell membranes of target cells, such as lung epithelial cells and macrophages. The resulting nanosponges inherit the surface antigen profiles of the origin cells and serve as baits to bind to SARS-CoV-2. This binding interaction blocks viral entry and inhibits viral infection (Zhang *et al.*, 2020).

polymeric nanoparticles in the neutralization of SARS-CoV-2.

Zhang et al. (2020) also showed that the microscopic sponge of human macrophages is more appropriate due to its ability to act both at the beginning and at the end of virus infection, although the nanosponge inactivation of human macrophages and nanosponge of human lung epithelial cells type II were similar for application in SARS-CoV-2. These researches stress the importance of conducting studies with longer time intervals than 3 days to identify whether nanosponges do not lose their efficiency or if they do not cause any damage to the body.

The use of cellular nanosponges for the treatment of SARS-CoV-2 infection requires additional validation in appropriate animal models, which promotes a discussion on the need for future research on clinical trials in humans, in order to verify whether or not it will present possible side effects. In addition, optimizing the main formulation can further improve the antiviral efficacy of these nanosponges. Thus, these small sponges serve to develop a new technology for the population, in order to protect the community from the new coronavirus.

CONCLUSIONS

The present study aimed to identify the antiviral activity of poly nanosponges (lactic acid- (PLGA) in the neutralization of SARS-CoV-2 through a literature review.

Based on the consulted researches, it was evidenced a virucida effect of poly nanosponges (lactic-coglycolic acid) against SARS-CoV-2 by approximately 90%, since the superficial polymeric structure of PLGA binds to the Spike spicule of the coronavírus. This binding prevents the spicule from interacting with the cell membrane, preventing the vírus entry into the cell.

REFERENCES

Ajinkya K, Prakash K, Vishal P. **Scaffold based drug delivery system: A special emphasis on nanosponges**. International Journal of Pharmaceuticas and Drug Analysis. 2015;3(4):98-104.

Astuti I, Ysrafil Y. **Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2): An overview of viral structure and host response.** Diabetes & Metabolic Syndrome: Clinical Research & Reviews. 2020;14(4):407-412.

Bezawada S, Charanjitha, Reddy VN, Naveen A. **Nanosponges: A concise review for emerging trends**. International Journal of Pharmaceutical Research and Biomedical Analysis. 2014;3(1):1-6.

Bilal M, Munir H, Nazir MS, Iqbal HM. **Persistence, transmission, and infectivity of SARS-CoV-2 in inanimate environments**. Case Studies in Chemical and Environmental Engineering. 2020;14(1):13-16.

Bhushan B. **Biomimetics: lessons from nature–an overview**. Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences. 2009;367:1445-1486.

Centers for Disease Control and Prevention. **"Coronavirus disease 2019 (COVID-19): situation summary". [Internet]**. 2020. [Cited 2020 Semptember 17]. Available from: https://www.cdc.gov/coronavirus/2019-ncov/cases-updates/summary.html.

Champion JA, Katare, YK, Mitragotri S. **Particle shape: a new design parameter for micro-and nanoscale drug delivery carriers**. Journal of controlled release. 2007;121(1-2):3-9.

Cui J, Li F, Shi Z. **Origin and evolution of pathogenic coronaviruses**. Nature Reviews Microbiology. 2019;17(3):181-192.

Dumitru AC, Espinosa FM, Garcia R, Foschi G, Tortorella S, Valle F, et al. **In situ nanomechanical characterization of the early stages of swelling and degradation of a biodegradable polymer. Nanoscale**. 2015;7(12):5403-5410.

Egawa N, Koshikawa N, Tomari T, Nabeshima K, Isobe T, Seiki M. **Membrane type 1 Matrix Metalloproteinase (MT1-MMP/MMP-14) Cleaves and Releases a 22-kDa Extracellular Matrix Metalloproteinase Inducer (EMMPRIN) Fragment from Tumor Cells**. Journal of Biological Chemistry. 2006;281(49):37576-37585.

Erbetta CD, Alves RJ, Resende JM, Freitas RF, Sousa RG. **Synthesis and characterization of poly (D,L-lactide-co-glycolide) copolymer**. Journal of Biomaterials and Nanobiotechnology. 2012;3(2):208-225.

Florindo HF, Kleiner R, Vaskovich-Koubi D, Acúrcio RC, Carreira B, Yeini E, Tiram G, Liubomirski Y, Satchi-Fainaro R. **Immune-mediated approaches against COVID-19**. Nature Nanotechnology. 2020;15(8):630-645.

Germolec DR, Shipkowski KA, Frawley RP, Evans E. **Markers of Inflammation**. In: DeWitt J, Rockwell C, Bowman C, eds. Immunotoxicity Testing. Durham: Humana Press; 2018. p. 57-79.

Gurunathan S, Qasim M, Choi Y, Tae Do J, Park C, Hong K, et al. **Antiviral Potential of Nanoparticles—Can Nanoparticles Fight Against Coronaviruses?**. Nanomaterials. 2020;10(9):1645-1674.

Hamidzadeh K, Christensen SM, Dalby E, Chandrasekaran P, Mosser DM. **Macrophages and the recovery from acute and chronic inflammation**. Annual review of physiology. 2017;79:567-592.

Hogan MC. **Respiration**. In: McGinley M, Cleveland CJ, eds. Encyclopedia of Earth. Washington: National Council for Science and the Environment, 2011.

International Committee on Taxonomy of Viruses (ICTV). [Internet]. 2019. [Cited 2020 September 17]. Available from: https://ictv.global/taxonomy/.

Jahagirdar PS, Gupta PK, Kulkarni SP, Devarajan PV. **Polymeric curcumin nanoparticles by a facile in situ method for macrophage targeted delivery**. Bioengineering & Translational Medicine. 2019;4(1):141-151.

Kamaly N, Yameen B, Wu J, Farokhzad OC. **Degradable controlled-release polymers and polymeric nanoparticles: mechanisms of controlling drug release**. Chemical reviews. 2016;116(4):2602-2663.

Kolate A, Kore G, Lesimple P, Baradia D, Patil S, Hanrahan JW, et al. **Polymer assisted entrapment of netilmicin in PLGA nanoparticles for sustained antibacterial activity**. Journal of Microencapsulation. 2015;32(1):61-74.

Letko M, Marzi A, Munster V. **Functional assessment of cell entry and receptor usage for SARS-CoV-2 and other lineage B betacoronaviruses**. Nature microbiology. 2020;5(4):562-569.

Mir M, Ahmed N, Rehman AU. **Recent applications of PLGA based nanostructures in drug delivery**. Colloids and Surfaces B: Biointerfaces. 2017;159:217-231.

Motta AC, Duek EA. **Síntese, Caracterização e Degradação "in vitro" do poli(L-ácido láctico-co-ácido glicólico)**. Revista Matéria. 2006;11(3):340-350.

Nieto-Torres JL, DeDiego ML, Álvarez E, Jiménez-Guardeño JM, Regla-Nava JA, Llorente M, et al. **Subcellular location and topology of severe acute respiratory syndrome coronavirus envelope protein**. Virology. 2011;415(2):69-82.

Patil MD. **Application of Nanotechnology in Covid-19**. Aayushi International Interdisciplinary Research Journal (AIIRJ). 2020;8(7):46-53.

Pendyala B, Patras A, Pokharel B, D'Sousa D. **Genomic modeling as an approach to identify surrogates for use in experimental validation of SARS-CoV-2 and HuNoV inactivation by UV-C treatment**. Frontiers in Microbiology. 2020;11:1-9.

Pourhajibagher M, Azimi M, Haddadi-Asl V, Ahmadi H, Gholamzad M, Ghorbanpour S, et al. **Robust antimicrobial photodynamic therapy with curcumin-poly (lactic-co-glycolic acid) nanoparticles against COVID-19: A preliminary in vitro study in Vero cell line as a model**. Photodiagnosis and Photodynamic Therapy. 2021;34: 102286-102298.

Rotello VM, ed. **Nanoparticles: building blocks for nanotechnology**. Amherst: Springer Science & Business Media; 2004.

Santos RM, Fialho SL. **Nanopartículas**. Biotecnologia Ciência e Desenvolvimento. 2007;37:52-59.

Schulz PA. **Nanomateriais e a interface entre nanotecnologia e ambiente. Vigilância Sanitária em Debate: Sociedade, Ciência & Tecnologia**. 2013;1(4):53-58.

Sharma S, Parmar A, Kori S, Sandhir R. **PLGA-based nanoparticles: A new paradigm in biomedical applications**. Trends in Analytical Chemistry. 2016;80:30-40.

Shereen MA, Khan S, Kazmi A, Bashir N, Siddique R. **COVID-19 infection: Origin, transmission, and characteristics of human coronaviruses**. Journal of advanced research. 2020;24:91–98.

Tan Q, He L, Meng X, Wang W, Pan H, Yin W, et al. **Macrophage biomimetic nanocarriers for anti-inflammation and targeted antiviral treatment in COVID-19**. Journal of nanobiotechnology. 2021;19(1):1-16.

Vazquez-Munoz R, Lopez-Ribot JL. **Nanotechnology as an Alternative to Reduce the Spread of COVID-19**. Challenges. 2020;11(2):15-25.

Wang Y, Sun J, Zhu A, Zhao J, Zhao J. **Current understanding of middle east respiratory syndrome coronavirus infection in human and animal models**. Journal of thoracic disease. 2018;19(10):S2260-S2271.

Watanabe S, Alexander M, Misharin AV, Budinger GR. **The role of macrophages in the resolution of inflammation**. The Journal of clinical investigation. 2019;129(7):2619-2628.

World Health Organization. **Coronavirus disease (COVID-19) Pandemic. [Internet]**. 2020. [Cited 2020 Semptember 17]. Available from: https://www.who.int/emergencies/diseases/novel-coronavirus-2019.

Yuan Z, Gu X. **Preparation, characterization, and in vivo study of rhein-loaded poly (lactic-co-glycolic acid) nanoparticles for oral delivery**. Drug Design, Development and Therapy. 2015;9:2301-2309.

Zhang Q, Honko A, Zhou J, Gong H, Downs SN, Vasquez JH, et al. **Cellular Nanosponges Inhibit SARS-CoV-2 Infectivity**. Nano Letters. 2020;20(7):5570-5574.