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DEVELOPMENT OF NANOCAPSULES FOR THE TREATMENT OF SPINAL CORD INJURY

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INTRODUCTION

Spinal cord injury (SCI) is a serious condition that leads to sudden loss of motor, autonomic and sensory function. The tissue injury associated with SCI is determined by a cascade of pathophysiological events that cause cell death, axonal loss, myelin degradation, infiltration and activation of immune cells, disruption of the spinal cord blood barrier, and glial scar formation. Vascular changes also occur and subsequent edema, ischemia, and hypoxia, production of cytokines, free radicals and lipid peroxidation, disruption of ionic balance, and excitotoxicity. These events impair neural regeneration and restoration of motor function (ANWAR, 2016, p. 98; PRÜSS, 2017, p.1549; KRONER, 2019, p. 134370; ZIMMERMANN, 2021, p. 353).

Thus, SCI is considered a multifactorial disease that seriously reduces the patient's quality of life, and there is currently no treatment with the potential for rehabilitation. Therefore, it is extremely important to develop new therapeutic strategies in this area that prevent the increase of severity of tissue damage.

Recently, many techniques based on nanotechnology have been proposed to improve tissue regeneration. Above all, nanoparticles, have interesting properties in the field of regenerative medicine, such as an ability to increase drug bioavailability. Furthermore, they potentiate drug penetration, exhibit excellent biocompatibility and reproducible microscopic structure (NEJATI-KOSHKI, 2017, p.85; NICOLA, 2017, p.95; FIROUZI-AMANDI, 2018, p.773; REIS, 2018, p.785; NICOLA, 2019, p.748).

Several studies have provided clear evidence that nicotine, an alkaloid that constitutes the active principle of tobacco, has powerful pathophysiological effects on the body, through anti-inflammatory properties. Studies suggest that nicotine improves tissue healing, increasing vascularization by stimulating angiogenesis, decreasing inflammation around the injury site, and accelerating fibrogenesis (HOU, 2008, p. 983; KENNETH, 2011, p. 349; PONS, 2011, p. 3842; KIM, 2017, p. e0179982).

In this work, different oils have been used in the preparation of nanocapsules (NCs), and the objective is to encapsulate the nicotine for the treatment of SCI and to compare in terms of diameter,

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polydispersity index (PdI), zeta potential, and encapsulation efficiency (EE).

METHODOLOGY

The nanocapsules (NCs) were prepared using poly (lactic-co-glycolic acid) (PLGA) through the technique of interfacial polymer deposition, where the emulsions were formed by deposition of an oil phase (OP) on an aqueous phase (AP), under vigorous magnetic stirring for 10 min at 40°C. Following this, the excess solvent and water were evaporated in a rotary evaporator at 40°C.

The group I was composed of PLGA, grape seed oil, acetone, Span 80, and nicotine in OP and distilled water and Triton X100 in AP. Group II was formed by PLGA, castor oil, acetone, Span 80, and nicotine in OP and distilled water and Triton X100 in AP. Group III was formed by PLGA, copaiba oil, acetone, Span 80, and nicotine in OP and distilled water and Triton X100 in AP. Group IV was composed of PLGA, açaí oil, acetone, Span 80, and nicotine in OP and distilled water and Triton X100 in AP.

The average size of the nanocapsules and the respective polydispersity index (PdI) were determined by the dynamic light scattering method (ZetaSizer Nano ZS, Malvern Instruments, Worcestershire, UK).

The encapsulation efficiency of nicotine in the nanocapsules was determined by the Liquid Chromatograph - Mass Spectrometer (LC-2040C Plus, Nexera-i, Shimadzu, Europe, equipped with the Shimadzu Shim-pack 100 C18 column). The following parameters were used: temperature 50°C, injected volume 0,5 µL, mobile faze A water and 0,1% acid formic, mobile faze B acetonitrile and 0,1% acid formic, and a flux 0.3 mL/min.

The sample was diluted in 0,5 mL of methanol, filtered, and injected into the LC-MS system. For determining the encapsulation efficiency, the samples were filtered in microfiltration tubes composed of 0.1 µm pore membranes (MilliPore) by centrifugation

(Thermo Scientific, SL8R, São Paulo, Brazil) for 10 min at 6000 rpm, and the supernatants were collected. Nicotine content in the supernatants (free nicotine) was also determined by LC-MS. The encapsulation efficiency was calculated as follows:

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Encapsulation efficiency = (total nicotine content in formulation - free nicotine) × 100/total nicotine content in formulation

RESULTS AND DISCUSSION

The NCs of Group I exhibited an average diameter of 214.9 nm, PdI of 0.351, zeta potential of -47.5 mv, and EE of 15.88%. Group II presented the average values: diameter of 207.7 nm, PdI of 0.271 and zeta potential of -27.2 mv, and EE of 91.27%. The NCs of Group III exhibited the average values: diameter of 190.2 nm, PdI of 0.421, zeta potential of -35.5 mv, and EE of 37.3%. Group IV presented the average values: diameter of 121.4 nm, PdI of 0.418, zeta potential of -22.3 mv, and EE of 32.45%.

These results reveal that the nanocapsules of all the groups have a suitable size and polydispersion, and also present values of zeta potential that reveal that the charges present on the surface of the nanocapsules prevent coalescence between them, avoiding the agglutination, attesting to their stability (SCHAFFAZICK, 2003, p. 726; MORAES, 2010, p. 995).

The encapsulation efficiency is highly dependent on the composition of the capsule core. The characteristics of the drug encapsulated, the oil used, and the presence of surfactants may facilitate drug solubilization (SCHAFFAZICK, 2003, p. 726). In this study, the NCs of Group II showed high encapsulation efficiency (91.27% of the total added), which means that this is the amount of drug stored in the nanocapsule core.

FINAL CONSIDERATIONS

In this study, it was possible to produce PLGA nanocapsules containing nicotine that were satisfactorily characterized about to their zeta potential, diameter, polydispersity index. It was also possible to verify that the nanocapsules of Group II presented excellent encapsulation efficiency concerning the other groups. The next phase will be the evaluation of the NC release profile.

Keywords: spinal cord injury; nanocapsules; poly (lactic-co-glycolic acid); nicotine

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