Microfluidic-assisted preparation of solid lipid nanoparticles for the brain-delivery of biologicals

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Traumatic brain injury (TBI) is a comprehensive term that refers to several impairments triggered by trauma affecting the brain. The pathological manifestation of TBI involves first a physical injury, which results in increased permeability of the blood-brain barrier (BBB), and then a significant release of pro-inflammatory mediators, which leads to a severe neuroinflammation event. During this detrimental phenomenon, the breakdown of the BBB creates the opportunity for passively directing non-invasive drug delivery systems (DDSs) to the brain to administer neuro-protective and/or neuro-regenerative agents. Current scientific research indicates that neurotrophins are important for the repair of injured brain parenchyma, and more specifically, the brain-derived neurotrophic factor (BDNF) could lead to beneficial regenerative effects. In this field, our research group demonstrated the potential of using solid lipid nanoparticles (SLNs), produced by microfluidics (MF), as a biomolecule DDS. Here, BDNF-SLNs have been successfully obtained by MF technique. The production was conducted through the before-optimized method using a commercially available device with a miniaturized modified herringbone geometry. Characterization data about the produced BDNF-SLNs showed the proficiency of MF-based production; in fact, this method allowed to achieve good findings in terms of dimensional range (190.3 ± 10.1 nm), PDI (0.180 ± 0.023), and ζ-potential (– 39.2 ± 1.30 mV), resulting in an encapsulation efficiency % of the 40%. An in vitro BBB model was built with a monolayer of immortalized human endothelial cell line (hCMEC/D3), and after 3 hours of permeation across the cell monolayer the data demonstrated a slight increase in the P_{app} BDNF-SLNs in comparison with plain BDNF at the same concentration of growth factor. For evaluating the potential therapeutic benefits of BDNF-SLNs in a TBI-like condition, an in vitro microglial cell model was built using the N9 cell line. Treatments with BDNF-SLNs and plain BDNF at the same concentration, namely 25 ng/mL and 100 ng/mL, were performed 4 hours before the addition of lipopolysaccharide (LPS, 1 µg/mL) and LPS+ interferon γ (IFN-γ, 0,1 µg/mL) and 1 hour after the addition of LPS and LPS+INF-γ. Empty SLNs were used as control. At the end of experiment, the amount of nitrites as NO products was measured to evaluate the neuro-protective effect of BDNF. In addition, real-time PCR analysis was conducted to follow the production of pro-inflammatory agents. As a result, data showed that the formulation of BDNF-SLNs (100 ng/mL) was capable to reduce the iNOS activation resulting in less nitrite production in each tested condition. Moreover, it has been demonstrated that encapsulated BDNF had a substantially greater ability to block iNOS activation than plain BDNF. Regarding real-time PCR results, the samples in which the toxic insult was added 4 hours after treatment with BDNF-SLNs (100 ng/ml) showed complete elimination of TNF-α cDNA compared to empty SLNs and plain BDNF. Eventually, BDNF-SLNs (100 ng/mL) highlighted potential neuro-protective activity in a TBI-compromised condition, opening the way for further evaluation.