Abstract

Growth factors represent a group of significant substances in the metabolism of organisms. They are signaling molecules that control cell activity at the endocrine, paracrine, or autocrine levels. They act as key mediators binding to cell receptors, triggering a cascade of reactions leading to the regulation of genetic transcription in the cell nucleus and stimulation of cellular response. Growth factors influence various physiological functions such as cell proliferation, cell differentiation, and tissue healing.

The utilization of growth factors is evident, for example, in regenerative medicine. For a similar purpose, research has been initiated to prepare the growth factor TGF-β3 with the possibility of attaching it to a polymeric carrier using a coiled-coil tag. This work focuses on the recombinant production of TGF-β3 – its analog with a latency-associated peptide (LAP) – and the application of techniques applicable to the intention of this work, specifically, attaching the protein to a polymeric carrier based on amino acids.

Given the structural complexity with which growth factors are physiologically released from cells, the preparation of growth factors with a coiled-coil tag *in vitro* represents an unexplored challenge in the field of recombinant protein expression. In our HEK293T cell line expression system, it was possible to prepare the latent form of LAP-TGF-β3, whose biological activity was verified on rat mesenchymal stem cells. Several constructs of the growth factor with a coiled-coil tag theoretically enabling its controlled release from the polymeric carrier into the culture medium were designed and successfully cloned. As part of the experimental work, the potential of sortase A was also discovered as an alternative method for preparing the desired protein.