EVALUATING THE STABILITY OF PLASMIDS WITH BIOTECHNOLOGICAL POTENTIAL FUNCTION

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Introduction and objectives: Plasmids are DNA molecules extrachromosomal double tape found in various taxons. They predominantly present under the supercoiled form, but they can assume other conformations as open circular, linear and relaxed, which directly influence their stability and applicability. Today the plasmid DNA (pDNA) has been studied as vector used in gene therapy and in DNA vaccines, using biotechnology as an ally. The aim of this study was to evaluate the stability of three pDNAs: pVAX1, pVAX1LacZ and MSPpVAX1 (encoding MSP gene - major surface protease of Trypanosoma brucei), when subjected to different temperatures during different time intervals. Materials and Methods: Competent E. coli DH5-α was produced, transformed with the pDNAs studied and subjected to different temperatures (-80, -20, 4, 22, 37 and 42°C) from 5 to 60 days. Subsequently, the transformation efficiency and different isoforms of pDNAs were carry out by agarose gel electrophoresis. Results and conclusions: The results show that plasmid conformation depends on temperature. The amount of supercoiled plasmids increase in the following order -80>-20>4>22>37>42°C. In addition, this order was not affected during the storage time, since after 60 days we found greatest amount of supercoiled plasmids in those samples that were under low temperature (-80°C). On the other hand, the plasmid stability and transformation efficiency decrease in the same order, because there were less supercoiled plasmids. We also found that the smaller plasmid (pVAX1) in all conditions used showed the best transformation efficiency. This result is contrary to what is reported in literature. Finally, we found that even in -80°C all plasmids showed traces of degradation which increasing their instability and decreasing their efficiency transformation, thus reducing their biotechnological potential as a vector. This data are important because they can have an impact on production, storage and use of pDNAs therapies with DNA.

Key words: plasmid, stability, transformation.

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