SPECIFIC FEATURES OF PHARMACEUTICAL TECHNOLOGY IN THE PRODUCTION OF PROTEIN DRUGS

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Introduction. Maintaining the structure of protein and peptide drugs has become one of the most important goals of scientists in recent decades. Cold and thermal denaturation conditions, lyophilization and freeze drying, different pH conditions, concentrations, ionic strength, environmental agitation, the interaction between the surface of liquid and air as well as liquid and solid, and even the architectural structure of storage containers are among the factors that affect the stability of these therapeutic biomacromolecules. The use of genetic engineering, side-directed mutagenesis, fusion strategies, solvent engineering, the addition of various preservatives, surfactants, and additives are some of the solutions to overcome these problems.

The aim of the study. To review the types of stress that lead to instabilities of different proteins used in pharmaceutics including regulatory proteins, antibodies, and antibody-drug conjugates, and then all the methods for fighting these stresses will be reviewed.

Methods of research. The literature on analytical methods used to detect instabilities, mainly changes in their primary structures and higher order structures, is reviewed.

Main results. The causes of instability include: physical instability (temperature-induced instability, cold denaturation, photo-induced instability, agitation-induced instability); chemical instability (hydrolysis, oxidation, disulfide exchanges, deamidation, conjugation-induced instability).

In many cases of the delivery of proteins and peptides for research use only, the powdered/dried form of proteins is preferable to their soluble form. In this delivery route, there is no protein hydrolysis pathways, in-solution decomposition, or air-water interactions caused by the agitation of the protein solution and environmental pH changes. Nevertheless, for the treatment sector and therapeutic usage, the powder form of protein drugs may cause many mistakes and consequences for patients, including mistakes in determining the correct dose of the drug, using solutions that may not have the required standard, and the need to send approved solvents from the companies. Thus, it is better to think through the solution form of protein and peptide drugs for therapeutic purposes [1]. Some of these strategies are related to the genetic design to produce more stable protein analogs, and some other paths are connected to protein structure and solvent engineering. The changes in genetics and the surrounding environment of medicinal proteins can be done with various goals such as increasing activity, increasing solubility, and protein stability.

Some of these approaches are related to the primary protein sequence (genetic changes) and many of these strategies are linked to protein changes after production and purification; the latter class usually has been divided into two main sub-classes: non-covalent and covalent modifications. These include techniques such as Genetic Engineering: Protein Analogs [2], Site-Directed Mutagenesis [3], Fusion Strategies [4], Protein-Polymer Conjugates [5], Linker Chemistry [6], Acylation [7], Cyclization

[8], Nanoparticles, Double-Blade Sword [9], Formulation: Solvent Engineering Pathways [10] and Choice of Container [11].

Conclusions. Looking at the sales share of protein and peptide drugs in recent years and their fast-growing trend, it is not far from expected that these macromolecules will cover most of the future therapeutic markets. Considering the positive features of this class of drugs compared to conventional chemical-synthetic drugs, such as high specificity, reasonable biological lifetime and low toxicity in the body, as well as the need for very low doses to create therapeutic responses, large pharmaceutical companies are willing to invest in this field. However, the problem with these drugs, since the beginning of their arrival, is their instability in different environmental conditions. For some therapeutic proteins, the binding of chemical molecules such as fatty acids has been a good option to increase their stability. Similar to the new approach used to produce antibody drug-conjugated, it is expected that a similar strategy will be adopted for some other therapeutic macromolecules such as DNA or RNA molecules in the future. Nowadays, most of the containers for storing therapeutic proteins and peptides are made of borosilicate glasses, which bring challenges such as the crushing of microscopic pieces of them in the solution and diverse interactions of proteins with their surfaces. Until now, a huge part of the application of nanoparticles and polymers has been used in the direction of drug (protein/peptides) delivery, however, in the field of engineering containers for medicinal proteins, no coherent effort has been made.

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