Intermediate synthetic scientific report
On the implementation of the project
NEW POLYMER / PEPTIDE HYBRID HYDROGELS AS INNOVATIVE PLATFORMS DESIGNED FOR CELL CULTURE APPLICATIONS

STAGE III / 2021

Sterilization of multicomponent systems,
Study of in vitro and in vivo properties,
Development of laboratory technology for the preparation of hybrid gel structures and use as a support/substrate for cell cultures (TRL4),
Dissemination of results

A1: Sterilization of multicomponent systems
A2: In vitro tests on hydrogels
A3: In vivo toxicology tests A4: Preparation of hybrid materials
A4: Selection of matrices that meet the requirements imposed by a device used for cell culture by correlating the results obtained from the physico-chemical and biological characterization.
A5: Development of laboratory technology for the integration of new hybrid materials as a support for cell cultures
A6: Project site dissemination, participation in scientific events, symposia, congresses

Results delivered on stage: LABORATORY TECHNOLOGY FOR OBTAINING NEW POLYMER/PEPTIDE HYBRID HYDROGELS AS INNOVATIVE PLATFORMS FOR CELL CULTURE APPLICATIONS

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Summary

Gels are a promising option for 3D cell cultures, constituting a basis for the adhesion and growth of cells as well as the transport of the nutrients they need. As structures supporting the development of cells, of natural or synthetic origin, composed of biocompatible materials, they allow their attachment (or seeding). Self-assembled gels are formed by the spontaneous aggregation of monomeric components into polymer-like fibrils through non-covalent interactions and the subsequent folding of these fibrils into an extended network that causes gelation. Self-assembly is a useful tool for the formation of hydrogels by structuring molecules through non-covalent bonds, generating networks that show similarities in organization with the cellular microenvironment. To be used as a support for cell cultures, the obtained hydrogels were sterilized beforehand. In this sense, several combined techniques were used: UV irradiation, ethanol washing of the prepared hydrogels, etc. For cell encapsulation, precursor solutions were also sterilized before hydrogel formation. This was achieved either by filtration (in the case of solutions with low viscosity), or by germicidal UV irradiation of the solution or the dry polymer. Evidencing the potential cytotoxic effects that a material can exert on cells is an important and decisive step in selecting the right materials for medical applications. In previous stages of the project, studies were carried out to evaluate the cytotoxicity of the obtained materials. Thus, they were incubated with primary Albino rabbit fibroblasts, and the cytotoxic effect was analyzed by measuring the metabolic activity of the cells, with the help of the MTT test and microscopic preparations in phase contrast and fluorescence. The cell viability data show that, although all the samples determined cell proliferation, this was different depending on the studied composition. The best cell viability was presented by the samples with agarose and fitagel in the composition. From the experimental data, it appears that the materials Agarose, FMOC-Lys-FMOC_Agarose, FMOC-Lys-FMOC_FMOC-Gly-Gly-Gly_Agarose are biocompatible, maintaining cell viability above 90% of the control value. The best results were obtained for the materials FMOC-Lys-FMOC_FMOC-Gly-Gly-Gly_Agarose for which cell viabilities over 90% were recorded at 72 hours of incubation with the cells. The analysis of the influence induced by the use of these gels on the hematological and immunological profile, as well as on the function and architecture of the liver and kidneys, demonstrates that the compositions used have a good biocompatibility in vivo, are non-toxic and safe for administration to rats, and suitable for biomedical applications. The evaluation of the physico-chemical properties (FTIR, SEM, thermal analysis, rheological studies) and the corroboration of the resulting information with those obtained from the evaluation of the biological characteristics (in vitro and in vivo tests) indicated that the matrix best meets the requirements imposed by a used device for cell culture variant FMOC-Lys-FMOC_FMOC-Gly-Gly-Gly_Agarose. On this matrix, the LABORATORY TECHNOLOGY FOR OBTAINING NEW POLYMER/PEPTIDE HYBRID HYDROGELS AS INNOVATIVE PLATFORMS INTENDED FOR CELL CULTURE APPLICATIONS, approved by the Internal Commission for evaluation and approval of results No. 3711/14.VII.2022.

Diseminaton
Papers:

1. A. P. Chiriac, E. Stoleru, I. Rosca, Al. Serban, L. E. Nita, A. G. Rusu, A. Ghilan, A. M. Macsim, L. Mititelu-Tartau; Development of a new polymer network system carrier of essential oils, Biomedicine & Pharmacotherapy 149, 112919, (2022) (**F.I. = 6,529**).

2. A. P. Chiriac, M. Asandulesa, I. Stoica, N. Tudorachi, A. G. Rusu, L. E. Nita, V. M. Chiriac, D. Timpu; Comparative study on the properties of a bio-based copolymacrolactone system; Polymer Testing 109, 107555, (2022) (**F.I. = 4,282**).
3. B.E.B Cretu, L. E. Nita, A.-M. Serban, A. G. Rusu, F. Doroftei, A. P. Chiriac, New Cryogels Based on Poly(Vinyl Alcohol) and a Copoly-Macrolactone System: I-Synthesis and Characterization, Nanomaterials 2022, 12(14), 2420; https://doi.org/10.3390/nano12142420 - 14 Jul 2022 (**F.I. = 4,282**).
4. A. Croitoriu, A. G. Rusu, A. Ghilan, M. Bercea, L.E. Nita, Aurica P. Chiriac, New FMOC-amino acids/peptides-based supramolecular gels obtained through co-assembly process: preparation and characterization, trimisa spre evaluare la Polymers. (**F.I. = 4,329**).

Presentation:
1. A. Croitoriu, A. M. Serban, A. P. Chiriac, A.G. Rusu, A. Ghilan, L.E. Nita, Hydrogels based on amino acids and gelling polymers; Polymers 2022 - New Trends in Polymer Science: Health of the Planet, Health of the People, Torino, Italia, Mai 25–27, 2022.
2. A. Croitoriu, L.E. Nita, A.G. Rusu, A. Ghilan, F. Doroftei, M. Bercea, A.P Chiriac; Hybrid hydrogels based on peptide; *International conference on rheology - Understanding the Viscoelastic Behavior of Materials – Progress and Challenges*; IASI, ROMANIA, Mai, 26, 2022