Institutul Național de Cercetare-Dezvoltare
în Domeniul Patologiei și Științelor Biomedicale
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Testing *in vitro* of Anti-inflammatory Potential in a Human Monocyte Cell Line

1. Service Objective

The service consists of evaluating the anti-inflammatory potential of one or more test samples, using a human monocyte cell line. The effect of the samples is analyzed by quantifying the cytokines released into the culture medium, at two relevant time points: 4 hours and 18 hours after treatment application. This approach allows characterization of the inflammatory/anti-inflammatory profile induced by the tested samples and its comparison with appropriate experimental controls.

2. Applications

The service can be used for:

- preliminary screening of compounds, extracts, formulations, or biomaterials with anti-inflammatory potential;
- evaluation of the cytokine response induced by samples under *in vitro* conditions;
- comparison of multiple concentrations or formulation variants;
- obtaining supporting data for research projects, experimental development, or product optimization.

3. Biological Material

The human monocyte cell line is used (CRL 9855^{TM}).
Cells are cultured in complete IMDM medium, supplemented with 10% fetal bovine serum, HT, and β-mercaptoethanol, under standard incubation conditions: 37 °C, 5% CO_2, humidified atmosphere.

3. Methodology

Cells are cultured in clear-bottom 24-well culture plates. Test samples are applied at concentrations established together with the client before the experiment begins.
Each experiment includes, as applicable: untreated control; solvent/vehicle control; positive control for activation of the inflammatory response; anti-inflammatory/comparative positive control, if available; cytotoxicity or cell viability control; test samples at one or more concentrations. Each experimental condition is performed in triplicate.
After treatment application, the culture supernatant is collected successively at 4 hours and 18 hours. Cytokines in the supernatant are determined using a multiplex technique, with a 6-plex kit. Results are expressed in pg/mL and interpreted comparatively with the controls included in the experiment. Examples of cytokines that can be analyzed: IL-1β, IL-4, IL-6, IL-8, TNF-α, IL-10, etc. (the cytokine set is established together with the client).

4. Deliverables

Final research report including a description of the methodology, raw data expressed in pg/mL at the 2 analyzed time points (4h, 18h), calibration curves for each molecule, and conclusions regarding the cytokine profile of the tested sample(s).

5. Estimated Duration

Approximately 15-20 working days from the start of the experiment, including: cell expansion time, treatment, supernatant collection, cytokine determination, and report writing.

6. Estimated Budget – 26 experimental scenarios with all controls included, performed in triplicate.

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Consumables subtotal	10475 lei
Labor subtotal	2000 lei
Total direct costs	**12475 lei**
Overhead (20%)	**2495 lei**
Profit (1%)	**124,75 lei**
Grand total	**15094,75 lei**

7. Terms

The test sample(s), as well as any additional requirements (concentrations, specific treatment conditions, etc.), will be established together with the client before the experiment begins.

The results are strictly for research/testing purposes *in vitro*.

To configure the study and provide a customized offer, information must be supplied regarding the sample type, the solvent, the proposed concentrations, and the testing objective.