PROJECT REVIEW

"Detection of Growth hormone by LC-MS Analysis"

Pr. G. Sanmartin, Dr. R. Gutierrez (University Pompeu Fabra, Spain)

One of the strategies for the detection of growth hormone (GH) administration is the identification of abnormal ratios between the main GH proteoforms in blood circulation. However, the high sequence similarity between them complicates a precise differentiation when relying on antibody-based quantification.

For unambiguous and precise protein identification, we will use liquid chromatography - mass spectrometry (LC-MS) analysis for the quantification of the 22 and 20 kDa GH proteoforms. Before, we will develop a sample preparation strategy for blood samples based on a novel type of (hydrogel) nanoparticles, for sample simplification and reduction of the blood matrix complexity. These nanoparticles have shown the capacity to simultaneously deplete blood from abundant proteins and enrich it in low abundance proteins through several types of particles and protocols, i.e. allowing for detection of low abundance proteins by LC-MS.

We will apply this methodology for the quantification of the 22 and 20 kDa proteoforms. We will further expand this strategy for the analysis of the 23 and 45 kDa proteoforms for which little information is available. The former showed a high concentration in post-exercise states. The latter could exist at higher amounts after administration of pharmaceutical GH. We will investigate their relevance within anti-doping analysis.

At the same time, we will purify blood extracellular microvesicles from blood samples to assess potential GH detection. These vesicles represent a blood sub-type of samples that have a much reduced matrix complexity as compared to blood. As such, the detection of low abundance proteins by LC-MS in this type of samples is significantly more accessible.