Session 2: New developments in flow cytometry for clinical diagnostic

Lectures

L.02.1

The phenotype of T regulatory cells as a predictor of the efficacy of Treg therapy

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T regulatory cells (Tregs) are considered a viable option in immunosuppressive treatment in the clinic. First promising clinical experiments and trials with clinical-grade Tregs cultured as advanced therapy medicinal product (ATMP) are completed already. We will present long-term results (up to 5 years follow up) of our trials with Tregs in type 1 diabetes and multiple sclerosis discussing metabolic and immune background of the patients.

L.02.2

Flow cytometry of extracellular vesicles – a new diagnostic tool?

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Flow cytometry is a multiparametric technology capable of characterizing single cells in suspension. However, currently, available flow cytometers are designed to detect also much smaller extracellular vesicles (EVs). Although signals originating from EVs are often on the borderline of the system sensitivity and partly overlap with the background noise, causing limitations and challenges with interpreting and reproducing EVs flow cytometry experiments, the growing list of evidence indicates the potential of this method as a diagnostic tool for many pathologies, including cancer and cardiovascular disorders. New technological advancements leading to more and more sensitive instrumentation dedicated to this type of analysis make the concept highly realistic. Current approaches allowing for flow cytometry analysis of EVs in different biological fluids with possible diagnostic relevance will be discussed in the presentation. The advantages and disadvantages of such a diagnostic tool will also be presented.

L.02.3

Myeloid-derived suppressor cells as a potential biomarker in cancer treatment and monitoring

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Myeloid-derived suppressor cells (MDSCs) represent a heterogeneous population of regulatory myeloid cells that rapidly expand under pathological conditions. According to their origin, they are divided into three main subsets: monocytic MDSCs (Mo-MDSCs), granulocytic or polymorphonuclear MDSCs (PMN-MDSCs), and early stage MDSCs (e-MDSCs). In cancer, the accumulation of MDCSs is inseparably related to the production of pro-inflammatory mediators by the tumor microenvironment (TME), which drives their suppressive activity.
Although MDSCs are detected in a high number in peripheral blood of cancer patients, their use as a diagnostic marker is limited as their blood level increases in premalignant states, as well as in infections and chronic inflammation. On the other hand, a meta-analysis showed that high levels of circulating MDSCs prior to treatment in most cancers correlate with worse survival of patients. Moreover, a growing list of studies show their potential role as predictors of response to the therapy. In melanoma patients the immunosuppressive activity of circulating MDSCs before and during the treatment with immune checkpoint inhibitors (ICI) could be used as indicator of the response to ICI therapy. In colorectal cancer, the level of MDSCs after surgery may be associated with tumor recurrence, while in prostate cancer, the level of circulating MDSCs after the treatment is dependent on the implemented therapy mode.

L.02.4

Utilization of HLA multimer assays in viral diagnostics

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Modern flow cytometry provides advanced HLA multimer assays that can be used in immune-monitoring virally infected individuals. The multimers consist of several HLA molecules targeted against a single viral protein that can bind to specific TCR receptors on the T cell surface. Moreover, multimers are fluorochrome-coded, which enables flow cytometry read-out.The HLA multimers can study anti-viral responses in immune-compromised individuals, like post-transplant or under immunosuppression therapy. If so, the humoral response could be reduced, and thus serological assays would be inefficient in confirming infection or anti-viral immunity. That could be the case for a tailored anti-viral therapy to treat patients upon their immune system's capacities.Some successful anti-viral HLA multimer assay examples in clinical practice have been proven recently. It was anti-CMV infection monitoring after BMT and anti-viral drug therapy reduction once the number of virally specific T cells reached the cut-off. Another was SARS-Cov-2 post-vaccine monitoring to stratify the risk of infection in immunodeficient patients.In summary, HLA multimer assays allow successful anti-viral monitoring of immunocompromised patients. It also provides a valuable tool for tailored therapy in virally infected individuals.