High hydrostatic pressure induces immunogenic cell death: implication for the design of dendritic cell-based antitumor immunotherapy

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The use of patient-derived dendritic cells (DCs) as a way how to elicit therapeutic immune response in patients suffering from cancer has been extensively investigated throughout the past decades. DCs are generated from peripheral blood monocytes, exposed to tumor antigens and then reintroduced into patients. Various methodological variations in each step of this process were tested by researchers in animal models as well as in humans in order to identify the optimal fnal product. One of the important step in this procedure is the selection of appropriate source of tumor antigens for the DC pulsation. The fact that the tumor specific rejection antigens are not generally defined and might differ inter as well as intra-patients, led us to the decision to use whole allogenic tumor cell lines as an optimal source of the tumor antigens. We tested various modalities of killing the tumor cells which fulfill the strict criteria compliant with the GMP (good manufacturing practice) such as safety and reproducibility and at the same time highest efficacy. Out of several physical and chemical modalities, high hydrostatic pressure (HHP) treated tumor cells displayed optimal characteristic. We first investigated the potential of HHP to induce immunogenic cell death (ICD) in human tumor cells which is important for the induction of antitumoral immune response. HHP induced the rapid expression of heat-shock proteins HSP70 and 90 and calreticulin, the main markers of ICD, on the cell surface and release of HMGB1 and ATP from the dying cells. The interaction of dendritic cells with HHP-treated tumor cells led to a more rapid rate of phagocytosis, upregulation of activation markers CD83, CD86 and HLA-DR and the release of proinflammatory cytokines such as interleukin IL-6, IL-12p70 and TNF-α by DC. DCs pulsed with tumor cells killed by HHP induced high numbers of tumor-specific T cells, while number of regulatory T cells which might negatively interfere with the efficient antitumoral response, remained low. We found that the key features of the endoplasmic reticulum stress-mediated apoptotic pathway, such as reactive oxygen species production, phosphorylation of the translation initiation factor eIF2α and activation of caspase-8, were activated by HHP treatment. Therefore, HHP acts as a reliable and potent inducer of ICD in human tumor cells [1,2]. The in vivo efficacy was then tested on animal model of prostate cancer: DC pulsed with HHP-treated tumor cells were as effective as docetaxel (DTX) in reducing prostate tumors in the orthotopic transgenic adenocarcinoma of the mouse prostate (TRAMP) model. Even if we did not observe any additive or synergic effects of chemoimmunotherapy on the tumor growth, only the combination of DTX and pulsed dendritic cells resulted in significantly lower proliferation of tumor detected by Ki67 staining in histological samples. In another clinically relevant setting, minimal residual tumor disease after surgery, administration of pulsed DC after the surgery of poorly immunogenic transplanted TRAMP-C2, as well as in immunogenic TC-1 tumors, reduced the growth of tumor recurrences [3].

The HHP-treatment of tumor cell lines was introduced into the manufacturing process of DCVAC, a dendritic cell based active immunotherapy, for the human use. The process was approved by local as well as international regulatory agencies (SÚKL, EMA, FDA) as GMP-compliant. The DCVAC has been tested in past 8 years by biotech company SOTIO in various stages of prostate, ovarian and lung cancer patients within the framework of clinical trials phase II. Encouraging results in ovarian cancer patients led to the Orphan drug status designation by EMA and FDA and the approval of a registration-enabling phase III study which is planned for coming next 5 years.

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