goal to instruct the generation of diagnostic tools and gather information for future immune intervention.

**Materials and Methods** Derivatives of the full-length HERV-H env gene were codon-optimized and synthesized based on annotated sequences. The wild type (WT) sequence was modified regarding the transmembrane-spanning domain including the cytoplasmic tail (TM+CT). For modified membrane tethered variants, the autologous TM+CT sequence was exchanged for a heterologous sequence. In addition, variants lacking the TM+CT or variants deprived of the complete transmembrane subunit were designed, resulting in the expression of a soluble secreted Env trimer or of the monomeric Env (SU). Expression of the HERV-H Env protein was analyzed by western blot and flow cytometry. Groups of 6 Balb/c mice were immunized with DNA vaccine constructs encoding the WT and modified HERV-H Env proteins on day 0 and 14 (prime), respectively, and boosted twice on day 42 and 70 with an adjuvanted formulation of a recombinant trimeric HERV-H Env protein. Antibody responses were monitored by ELISA against various HERV-H Env (trimer, SU and extracellular domain of TM).

**Results** Western blot and flow cytometry analysis showed proper expression of membrane-bound Env proteins with no significant enhancement of cell surface display for the TM modified protein. Antibody responses against all Env variants could be shown already after two DNA immunizations and were elevated after boosting with the recombinant Env protein. Except for the group immunized with the DNA vaccine encoding the SU, which showed significantly higher antibody titers compared to all others, no differences could be seen between the groups following the protein boost.

**Conclusions** The results prove the antigenicity and immunogenicity of the HERV-H Env protein and its derivatives. All engineered HERV-Env derivatives were able to prime Env specific antibody responses in Balb/c mice. Booster immunization with adjuvanted Env trimer yielded comparable antibody titers, except for SU, which showed superior responses in terms of magnitude. Data and reagents described herein provide a valuable foundation for the development of diagnostic tools for tumor stratification, antibody-based intervention as well as therapeutic vaccination strategies. 1. Zhang, M, Liang, JQ, Zheng, S. Expressional activation and functional roles of human endogenous retroviruses in cancers. Rev Med Virol. 2019; 29:e2025. https://doi.org/10.1002/rmv.2025

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**P03.08** NEOANTIGENIC VERSUS MULTI-ANTIGENIC PERSONALIZED B16 MELANOMA VACCINES COMPARISON ACCORDING TO ANTI-TUMOR T CELL RESPONSE INTENSITY

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**Background** Personalized tumor vaccines based on synthetic peptide neoantigens or those encoded in RNA-vector are successfully studied in phase 1 and 2 clinical trials in melanoma and glioblastoma patients.

**Objectives** In our study we compare immunogenicity of two personalized tumor vaccine platforms, one based on DNA-vector encoding tumor neoantigens and the other multiantigenic vaccine made of primary tumor tissue with included molecular immunoadjuvants.

**Materials and Methods** As a neo-antigenic tumor vaccine (Ad-B16) the recombinant adenovirus vector, encoding B16F10 melanoma mutant antigen, was used. A multi-antigenic tumor vaccine (MTV-B16) was made out of the B16F10 tumor tissue. PRR-agonistic molecular immunoadjuvants were included in the multiantigenic B16 tumor tissue-derived vaccine in order to activate antigen-presenting dendritic cells and repro-gram myeloid suppressors. The number of antigen-reactive IFNγ-secretory T effector and T effector memory cells was analyzed by ELISPOT. Serum antibodies specific to intracellular antigens of B16F10 melanoma cells were analyzed using ELISA, while FACS was applied to detect B16F10 surface antigens.

**Results** MTV-B16 vaccine show stronger immunogenicity than Ad-B16 as to generation of tumor-specific IFNγ-secretory T cells in the spleen of mice. After immunization with MTV-B16, up to 6500 IFNγ CD4 and 3500 IFNγ CD8 T-effector cells (per 1 mln T cells) were discovered versus 600 IFNγ CD4 and 650 IFNγ CD8 T-effector cells generated by Ad-B16. Both vaccines induced serum antibodies specifically recognizing B16F10 melanoma’s intracellular and cell surface antigens.

**Conclusions** A personalized multi-antigen tumor vaccine MTV-B16, made out of tumor tissue and completed with molecular adjuvants, induces significantly stronger tumor-specific Th1-type CD4 and CD8 T cell responses than those generated by the neoantigen Ad-B16 vaccine on the adenovirus vector platform.

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**P04 Precision medicine meets immunotherapy (immuno-monitoring)**

**P04.01 VOLATILE PROFILING USING AN ENOSE ALLOWS DIFFERENTIATION OF VOLATILE PHASES DERIVED FROM SERUM, DC, OR MLC CULTURE SUPERNATANTS FROM HEALTHY OR LEUKEMIC SAMPLES**


**Background** Volatile organic compounds (VOCs) reflect the metabolism in healthy and pathological conditions. They can be collected easily in a noninvasive matter, directly measured by electronical nose (eNose) and might qualify as a systemic tool to monitor biomarkers related to disease. 1. Myeloid leukemia blasts can be transformed into leukemia derived dendritic cells (DCleu) being able to improve (anti-leukemic) immune responses. 2. To profile the immunological changes in healthy