

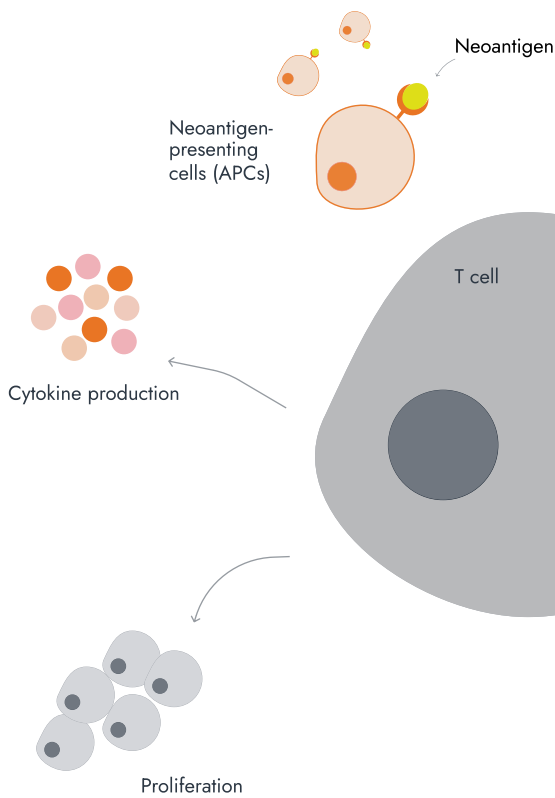
Antigen selection and validation

Experimental validation of neoantigens selected by differently tailored algorithms for vaccine development projects involves several key approaches to ensure their immunogenicity and effectiveness.

In-vitro

Neoantigens are tested using **T cell assays** (EliSpot/ FluoroSpot) to evaluate their ability to stimulate T cell responses. This means co-culturing neoantigen-presenting cells (APCs) with T cells to assess activation, proliferation, and cytokine production.

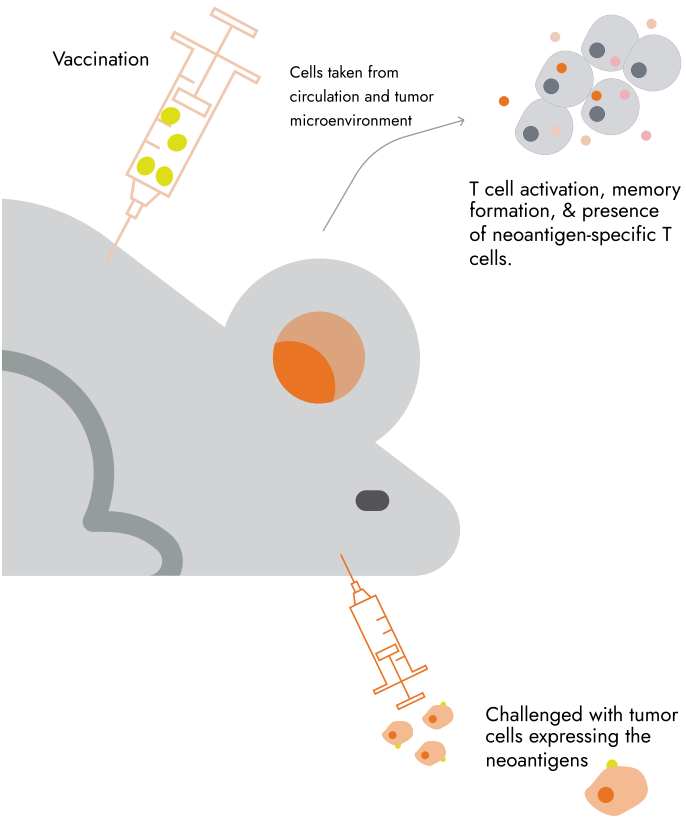
The data obtained may be complemented by **MHC binding studies**. The binding affinity of neoantigens to MHC molecules is measured using techniques such as peptide-MHC stabilization assays. Strong binding is indicative of potential immunogenicity.



In-vivo

Following vaccination in preclinical animal models, the immune response is analyzed by measuring T cell activation, memory formation, and the presence of neoantigen-specific T cells in the circulation and tumor microenvironment.

This may include **preclinical tumor challenge experiments** where vaccinated animals are challenged with tumor cells expressing the neoantigens to assess the therapeutic or prophylactic efficacy of the vaccine and its ability to prevent tumor growth.



Specific neoantigens or platforms used for their identification that show promise in **preclinical models, early-phase clinical trials** are conducted to evaluate safety, tolerability, and immunogenicity in human subjects.

Through these experimental validations, researchers aim to **confirm the potential of selected neoantigens to elicit effective immune responses, paving the way for successful vaccine development against cancer**. Core assay to judge the success rate of selected neoantigens is EliSpot/FluoroSpot as it measures the quantity of specific T/B cells against the antigen at single timepoints but also over time (immune kinetics). For that you need to be sure that your assay is robust and validated to fit regulatory aspects too.