PEPperPRINT
A NEW DIVERSITY

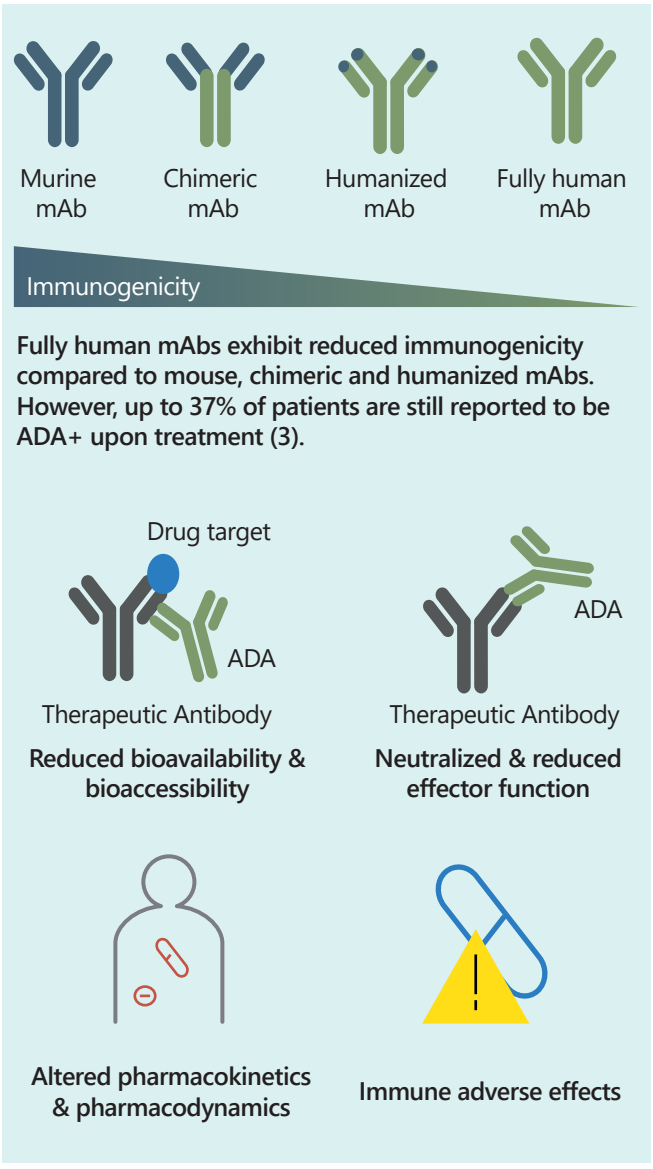
One-stop Solution for the Fingerprint Analysis of Immune Adverse Effects

Monitor pre-existing and drug- related antibody and T-cell responses for immunogenic adverse effects risk assessment. Identify immunogenic epitopes recognized by anti-drug antibodies (ADA) in a single assay.

Therapeutic protein products like monoclonal antibodies (mAb) have revolutionized the treatment of several diseases and disorders, including cancer, chronic autoimmune and inflammatory diseases (1). However, even the FDA-approved, fully human mAbs are reported to be highly immunogenic. This immunogenicity is associated with anti-drug antibodies (ADA) generation, which can neutralize drug efficacy by altering its pharmacokinetic and pharmacodynamic properties. Above all, ADAs cause several immune adverse effects in patients (1).

Discovering biomarkers that identify patients at risk is essential to design safer therapeutic protein drugs with lower rejection rates. The immunogenicity of e.g. mAbs can manifest in the form of different immune responses depending on the patient's T-cell and B-cell repertoires, cytokine milieu, and prior exposure of the immune system to proteins of similar structure. ADAs can be generated by T-cell-dependent or independent B-cell activation pathways. Identification of ADA epitopes in polyclonal Ab mixtures like sera is usually challenging due to high diversity. High-density peptide microarrays are a powerful tool to simultaneously screen tens of thousands of peptides against serum antibodies in a high-throughput context (2).

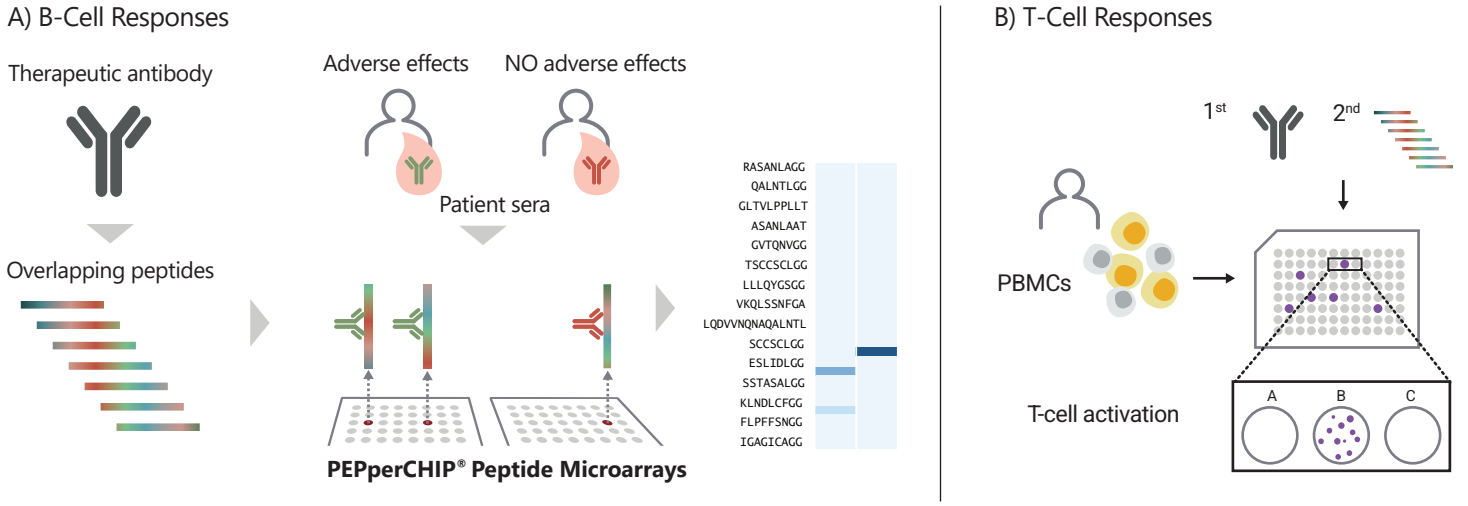
At PEPperPRINT, we provide the fingerprint immunogenicity analysis of your antibody or protein drug in a single assay with epitope mapping technology. Moreover, we provide T-cell services to determine the presence of mAb-reactive T-cells in patient PBMCs by ELISpot. This dual strategy enables comparison of the immune landscape among patients with and without adverse effects.



1) Vaisman-Mentesh A, *et al.* Front. Immunol. 11:1951 (2020)

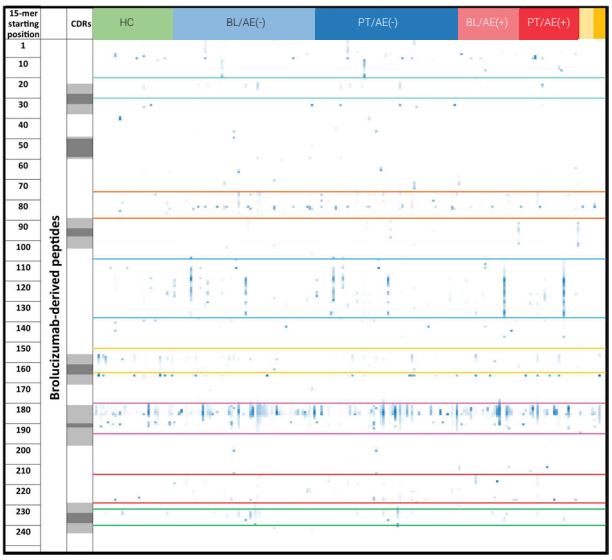
2) Heiss K, *et al.* J Proteome Res. Nov 6;19(11):4339-4354 (2020)

3) Agrawal S, *et al.* J Clin Pharmacol. Mar;57(3):394-400 (2017)



Exemplary workflow: A) To detect the anti-drug responses on the epitope level, serum or plasma samples from different patient groups are screened with PEPperCHIP® Peptide Microarrays displaying overlapping peptides of a therapeutic mAb or protein drug of interest. Identified hit peptides are presented as a heat map based on their reactivity, in addition to an immunoglobulin isotype/subtype analysis. B) ELISpot assays in PBMC cultures allow for direct-testing of T-cell antigenicity of a protein drug or mAb. For further analysis of epitope-specific T-cells, promising peptide candidates are synthesized and tested in ELISpot assays with patient PBMCs. The resulting response profiles can be compared across different patients.

Use Case



BL: Baseline
PT: Post-treatment
AE: Adverse effects
HC: Healthy controls

In 2019, Novartis launched brolucizumab, a single-chain variable fragment targeting VEGF-A for the treatment of neovascular age-related macular degeneration. In 2020, rare cases of retinal vasculitis and/or retinal vascular occlusion were reported, often in the first few months after drug administration. To identify immunogenic sites recognized by anti-brolucizumab ADAs, we generated a high-density peptide microarray by converting the brolucizumab sequence into a 15 amino acid peptides with a max. peptide-peptide overlap of 14 amino acids for high-resolution epitope data. The brolucizumab peptide microarrays were tested with the serum samples of different patient and control groups to identify IgG, IgA and IgM antibody responses on the epitope level. That way a class-switched, high-affinity immune response was observed, with several linear epitopes being recognized by ADA.

Figure derived from Karle, Anette C., et al. "Anti-brolucizumab immune response as one prerequisite for rare retinal vasculitis/retinal vascular occlusion adverse events" Sci Transl Med 2023.

Applications

- Monitoring of patient-specific anti-drug antibody and T-cell responses in clinical trials and phase IV studies.
- Assessment of treatment risks and immunogenic adverse effects.
- Identification of immunogenic B-cell and T-cell epitopes for development of safer antibody and protein drugs.
- Analysis of antibody profiles including immunoglobulin isotypes and subtypes.

Company Snapshot

PEPperPRINT is an innovative biotech company based in Heidelberg, Germany, and provides state-of-the-art products and research solutions for immunology and precision medicine. The company is focusing on infectious, autoimmune and chronic diseases as well as immuno- oncology.

PEPperPRINT is using a validated and trusted biomarker platform and holds a strong IP portfolio which enables extended services for strategic collaborations with world leading research centers (NCT, University Hospital Heidelberg, German Cancer Research Center) and qualified and contracted top pharma companies like Novartis, GSK, Boehringer Ingelheim, and many more.