DESCRIPTION

Empty baculovirus particles (BVPs) (empty viral capsids without genomes) can be used to measure polyspecificity of therapeutic antibody candidates in in vitro assays. This assesses the viability of an antibody as a therapy early on. They are provided as viral particles in phosphate buffered saline (PBS) at a concentration of 1 x $10^{12\text{-}13}$ pfu/mL. 100 µL is enough to assay up to 4 x 96-well plates at a final dilution of 1:200.

BACKGROUND

Some antibodies are able to recognize more than one antigen, which is referred to as polyspecificity (Van Regenmortel, M.H.V., 2014). Therapeutic antibodies have different half-lives in the body that can affect their therapeutic efficiency. One reason for fast clearance from the body is the polyspecificity of the antibody. An ELISA assay using non-specific binding to baculovirus particles (BVPs) of therapeutic antibodies showed correlation with the fast in vivo clearance of antibodies (Hötzel et al., 2012). Determining the polyspecificity of an antibody early on during therapeutic antibody development helps with decision making whether to pursue the antibody therapy further.

References:

Van Regenmortel, M.H., “Specificity, polyspecificity, and heterospecificity of antibody-antigen recognition”, J Mol Recognit, 2014. 27(11): p. 627-39.
Hötzel et al., “A strategy for risk mitigation of antibodies with fast clearance”, mAbs, 2012, 4:6, 753-760. Xu et al, “Addressing polyspecificity of antibodies selected from an in vitro yeast presentation system: a FACS-based, high-throughput selection and analytical tool.”, 2013, Protein Engineering, Design & Selection, 26:10, pp. 663-670.

FORMULATION

PBS

STORAGE

-80ºC

Note: 1:100 – 1:200 dilution is recommended. The actual dilution used must be determined empirically to meet end user’s needs.

PROTOCOL

1. Make appropriate dilutions of BVP with 50 mM sodium carbonate (pH 9.6).
2. Add 50 uL to each well. Incubate 16-24 h at 4ºC.
3. Aspirate the unbound BVPs from the wells and perform the rest of the steps at room temperature
4. Add 100 µL of blocking buffer (PBS with 0.5% BSA) and incubate for 1 hr.
5. Wash 3 times with 100 µL of PBS.
6. Add 50 µL of 1 µM primary antibody and incubate for 1 hr.
7. Wash 6 times with 100 µL of PBS.
8. Add 50 µL of anti-Human-IgG-HRP and incubate for 1 hr.
9. Wash 6 times with 100 µL of PBS.
10. Add 50 µL of TMB substrate and incubate for 10-15 min.
11. Stop the reaction with 50 µL of 2M sulfuric acid.
12. Read absorbance at 450 nm.
13. BVP score is determined by normalizing absorbance by control wells with no test antibody.