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Optimization of an anti-poly(ethylene glycol) (anti-PEG) cell-based capture system to quantify PEG and PEGylated molecules

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S ensitive determination of the pharmacokinetics of PEGylated molecules can accelerate the process of drug development. Here, we combined different anti-PEG Fab expressing 293T cells as capture cells (293T/3.3, 293T/6.3, and 293T/15-2b cells) with four detective anti-PEG antibodies (3.3, 6.3, 7A4, or 15-2b) to optimize an anti-PEG cell-based sandwich ELISA. Then, we quantified free PEG (mPEG2K-NH₂ and mPEG_{5K}-NH₂) or PEG-conjugated small molecules (mPEG_{5K}-biotin and mPEG5K-NIR797), proteins (PegIntron and Pegasys), and nanoparticles (Liposomal-Doxorubicin and quantum-dots). The combination of 293T/15-2b cells and the 7A4 detection antibody showed best sensitivity for free PEG, PEG-like molecules, and PEGylated proteins with detection at ng mL⁻¹ levels. On the other hand, 293T/3.3 cells combined with the 15-2b antibody had the highest sensitivity for quantifying Lipo-Dox at 2 ng mL-1. All three types of anti-PEG cells combined with the 15-2b antibody had high sensitivity for quantification down to 7 pM. These results suggest that the combination of 293T/15-2b cells and 7A4 detection antibody is the optimal pair for sensitive quantification of free PEG, PEG-like molecules, and PEGylated proteins, whereas the 293T/3.3 cells combined with 15-2b are more suitable for quantifying PEGylated nanoparticles. The optimized anti-PEG cell-based sandwich ELISA can provide a sensitive, precise, and convenient tool for the quantification of a range of PEGylated molecules.

Biography

Wen-Wei Lin received BS degree in 2009 from Kaohsiung Medical University, Taiwan, with a major in Biomedical Science and Environmental Biology. He received MSc degree in 2011 from the Graduate Institute of Oral Biology at National Taiwan University. He is currently a PhD student at the Institute of Biomedical Sciences in National Sun Yat-sen University, working in the laboratory of Prof. Tian-Lu Cheng for development of antibody drugs. His research interest is antibody engineering, targeted therapy of cancer and drug discovery.

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