GTP Bioways
CDMO
Continuum for biologics

Case Study

Purification of an ADC to remove the residual free drug

The Challenge

Two directions were followed to reduce the residual free drug level: optimization of the coupling procedure, and development of a specific TFF step.

Purification of an ADC to remove the residual free drug

Context & Challenge

Antibody-Drug Conjugates (ADC) are a class of complex molecules made of three components: an antibody (mAb), a linker, and a drug (or payload). The antibody allows specific targeting of the ADC while the drug, which is the active component, can fill its role. ADCs are indeed mostly designed to destroy exclusively cancer cells leaving healthy cells unaffected. ADC development and manufacturing is challenging as it requires (1) the development of production and purification processes for the mAb, the linker, and the drug, (2) the development of the coupling procedure of the drug to the mAb, and (3) the purification of the ADC.

In the present study, a major difficulty was encountered when purifying the ADC. Following coupling of the drug to the mAb, the free drug that did not react must be removed. Let us remember that it is a cytotoxic compound, and, without any driver, it would kill any cell and not only cancer cells when injected to a patient. In this case, the drug would not be efficiently washed away using the standard procedure at GTP Bioways.

GTP Bioways' Solution

Two directions were followed to reduce the residual free drug level: (1) optimisation of the coupling procedure, and (2) development of a specific Tangential Flow Filtration (TFF) step.

The optimisation of the coupling procedure was achieved following a Design of Experiments (DoE) approach. Buffers, reaction times, and concentrations of the two reagents were varied to determine the most appropriate condition. This step allowed to reduce the level of free drug after conjugation and before the purification step (Fig. 1).

A TFF step dedicated to the efficient removal of the drug was developed. Combinations of different membranes and buffers were systematically tested, and the best condition was picked up. Then the decrease in free drug was modelled to determine the diafiltration volume sufficient to reach the lowest residual free drug level possible (Fig. 2).

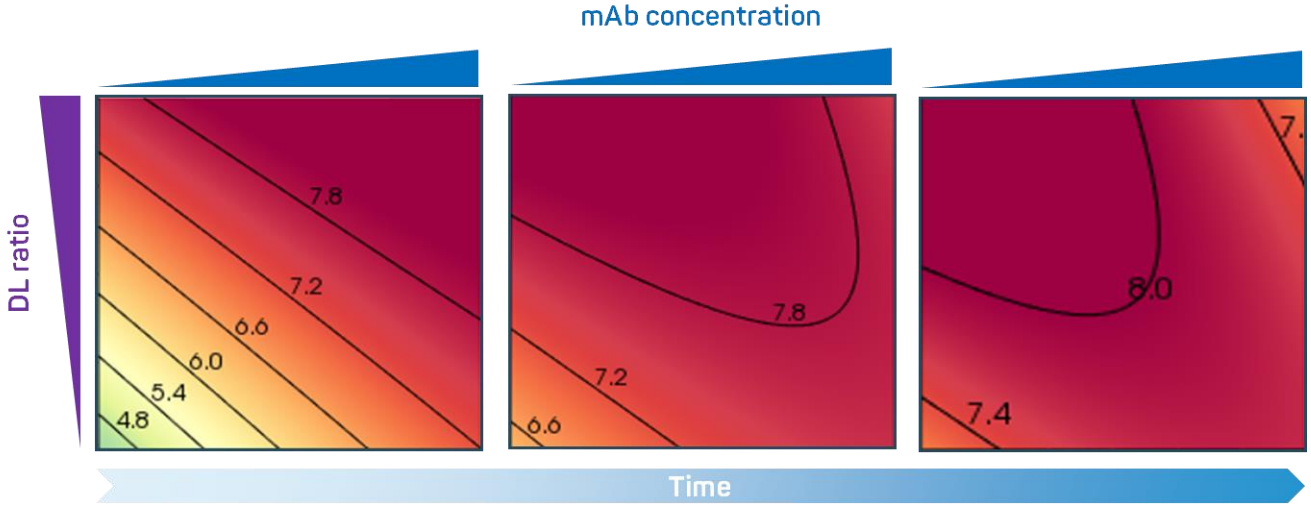

Figure 1. A DoE approach was setup to determine the optimal conjugation conditions. The Drug-Linker (DL) ratio, the antibody (mAb) concentration, pH, and time were three parameters that were modulated. The lines on the maps represent various buffer pH values.

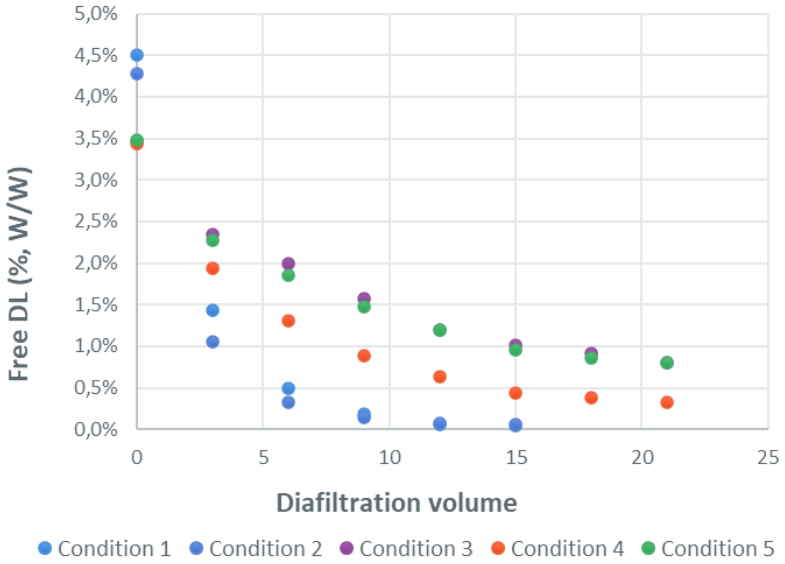

Figure 2. A Modelling Approach Optimising the Diafiltration Volume to Minimize Residual Free Drug Levels

> "Removing the drug linker can be challenging and relies on how it interacts with the membranes employed during the purification process. Through research, perseverance, and teamwork, we have succeeded in winning this race against time."
>
> Perrine BAZAUGOUR, DSP technician

Our client's success

The combination of the two approaches led to a reduction of residual free drug three times below the specification set by our customer and allowed the initiation of pre-clinical and clinical studies in due time. As always, the successful achievement in a short amount of time lies in a strong cooperative effort of the USP, DSP, and QC teams.