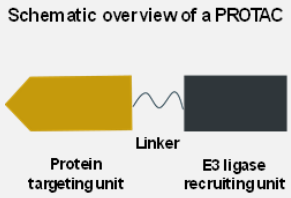
PROxAbs: Next generation PROTACs to access difficult targets

A modular kit using antibodies overcomes current PROTAC limitations

Conventional drug discovery focuses on regulating protein function by blocking specific target sites. Unfortunately, not all disease-related proteins are targetable due to a lack of accessibility. To overcome this problem, targeted protein degradation (TPD) has gained increasing interest in recent years. With this approach, protein targets could be degraded in a short time, thus providing a powerful and interesting tool for the development of new drugs.



The PROTAC technology harnesses the cell's own machinery to selectively destroy disease-associated target proteins.

Currently there are at least 20 ongoing clinical trials investigating the effect of PROTACs in various malignancies.[1]

Several degradants have been explored so far and one of them is PROteolysis Targeting Chimeras (PROTAC).[2] PROTACs are heterobifunctional molecules consisting of three components – a protein of interest (POI) targeting moiety, an E3 ligase recruiting moiety, and a linker connecting both moieties.

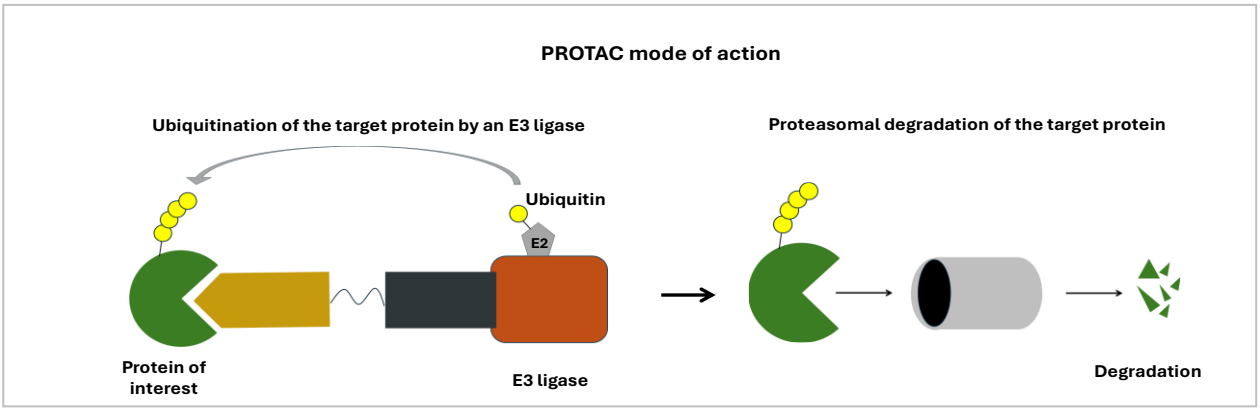


Figure 1. *Schematic overview of targeted protein degradation with PROTACs.*

PROTACs function by bringing a POI and a E3 ligase into proximity and initiating the ubiquitination of the POI by the E3 ligase machinery, resulting in a degradation of the POI by the proteasome **(Figure 1)**. The two most commonly used E3 ligases for targeted protein degradation by PROTACs are the Cereblon (CRBN) and the Von Hippel-Lindau tumor suppressor (VHL).

Although PROTACs have been shown to be effective, their use faces challenges such as a short half-life time, low cell permeability and large molecular size.[3-5] Another important limitation is the lack of tissue specificity as PROTACs interact with the broadly expressed CRBN and VHL E3 ligases. And finally, the covalent coupling technology currently required for the antibody-PROTAC complex relies on linker sequences that are difficult to attach to PROTACs and negatively impact functionality by the increasing chemical complexity. These disadvantages of PROTACs need to be urgently addressed to increase the use of antibody-PROTAC complexes in various therapeutic areas.

Improved PROTAC applicability through E3 ligase specific antibody and a fast & easy complexation

A new modality for specifying TPD is the combination of antibodies with TPD modalities. Hendrik Schneider and colleagues (Merck KGaA) explored a novel platform for linking PROTACs to bispecific antibodies.[6] In collaboration with YUMAB GmbH, Schneider and co-workers developed a single-domain camelid antibody, specific for the VHL E3 ligase recruiting subunit (anti-VH032) of PROTACs that was genetically fused to an existing therapeutic antibody thereby creating a bispecific antibody.

YUMAB has extensive expertise in the development of camelid derived single domain (VHH) antibodies which, due to their small size and elongated CDR3 loop, are capable of interacting with difficult to access epitopes.

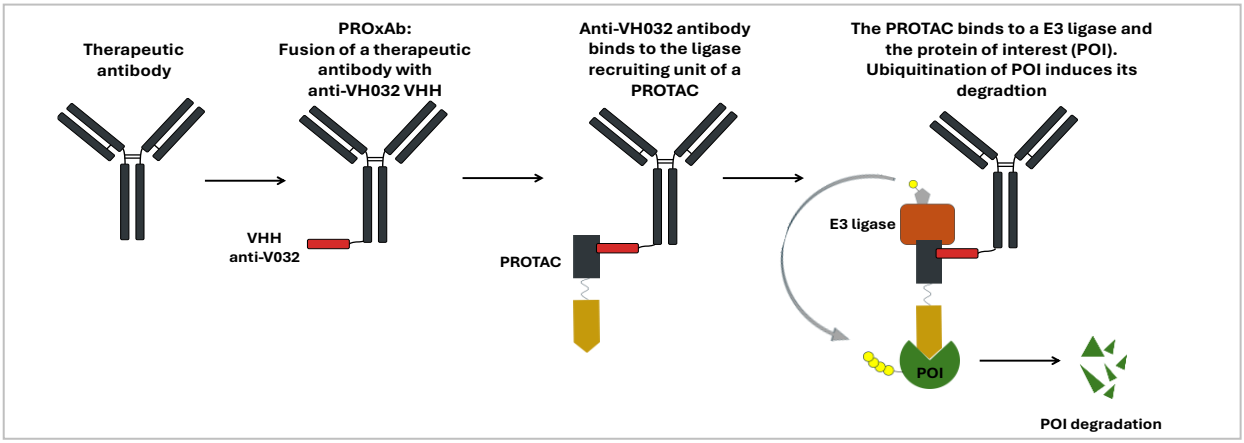


Figure 2. *Interplay of PROxAb shuttle and a PROTAC*

Schneider et al. used their non-covalent coupling technology to form PROTAC-Antibody shuttles (PROxAb) by complexing the bispecific antibody (anti-VH032 + therapeutic antibody) with the VHL-binding portion of the PROTAC **(Figure 2)**. This, in a couple of minutes generated complex internalizes into cells and shows in vivo a prolonged half-life in comparison to PROTAC alone. The non-covalent coupling of the PROTAC has the great advantage of not requiring any linker chemistry or bioconjugation techniques. Since the biology of the PROxAb shuttle remains intact, the new construct provides a plug-and-play platform with lower complexity compared to other antibody-PROTAC conjugates.

This innovative PROxAb approach clearly improves efficacy and therapeutic applicability of PROTACs by ensuring specificity, increasing half-life in vivo and simple producibility.

Just like antibody-drug conjugates (ADC) that enable targeted drug delivery – with 13 currently approved compounds on the market[7] – PROxAb could be used as complementary therapy for many difficult to access targets in a broad range of diseases.

REFERENCES

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