

NIH COMMON FUND HIGH-RISK HIGH-REWARD RESEARCH SYMPOSIUM

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SPEAKER ABSTRACTS

Single domain antibodies as tools to perturb protein interactions

Awardee: Hidde L Ploegh

Award: Pioneer Award

Awardee Institution: Whitehead Institute for Biomedical Research

The proposed use of single domain antibodies (VHHs) from camelid derived heavy chain only immunoglobulins was geared towards phenotypic screens in yeast and possibly other eukaryotes as a novel means of perturbing the host cell proteome. We have now conducted such phenotypic screens in yeast as a means of inhibiting pyruvate decarboxylase to redirect production of ethanol to that of other higher alcohols, with the first indications of success. We have also conducted phenotypic screens to identify VHHs capable of interfering with the influenza virus life cycle by cloning inducible versions of VHHs in lentiviral vectors, followed by exposure of the transductants to a lethal dose of virus. We thus identified VHHs capable of interfering with the function of flu NP, yielding a near complete blockade in virus replication. We have obtained co-crystals of the inhibitory VHH with recombinant NP, the structure of which will help us identify the essential functionally relevant features of NP that lead to its inhibition by the relevant VHHs. More generally, we have generated several VHHs that have assisted in the crystallization of otherwise difficult to crystallize proteins and thus helped arrive at their structures. This same pipeline was used to generate VHHs against proteins of immunological interest, ultimately with the goal of providing new imaging modalities to explore host-pathogen interactions. We can now image immune cells non-invasively using PET imaging. These approaches demonstrate the ease of manufacture and utility of camelid single domains in experimental approaches where conventional antibodies are less useful or fail altogether.