The power of chemoselectivity: Functional peptide- and protein-conjugates for extra- and intracellular targeting

Christian Hackenberger

FMP and HU Berlin, Germany

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Chemoselective reactions are essential synthetic tools employed in all areas of chemistry. In addition to their application in the total synthesis of various natural products, they have advanced research in the material and life sciences by allowing the conjugation of various biological molecules to polymers, nanoparticles and other materials as well as the straightforward chemical modification of proteins for biological and pharmacological purposes. Moreover, the development of chemoselective protein ligation strategies, spearheaded by the engineering of native chemical ligation[1] and expressed protein ligation[2] for the conjugation of two unprotected peptide or protein fragments from either synthetic or biological origin, have advanced the field of protein synthesis culminating in the impressive synthesis of naturally occurring posttranslationally modified proteins.[3] Another remarkable advancement in this area was the development of bioorthogonal reactions, which introduced the concept of performing chemical reactions in living organisms by modifying unnatural functional groups, so-called bioorthogonal reporters, present inside and outside of cells leading to the development of new probes to study extra- and intracellular pathways.[4]

In the Zervas Award lecture an overview is given on our recent investigations on the identification of new chemos-elective and bioorthogonal reactions for the synthesis and modification of functional peptides and proteins.[5] We apply these reactions to study functional consequences of naturally occurring posttranslational protein modifications (PTMs), in particular to study phosphorylation and O-GlcNAcylation events on the Alzheimer relevant Tau-protein.[6] Moreover, by utilizing chemoselective Staudinger reactions of azides with phosphites [7a,b,f] and phosphonites [7c,d,e] as well as the reaction of phosphites with electrophilic disulfides,[7g] we obtained phosphorylated Lys- and Cystein-peptides for to advance the proteomic analysis of these labile underexplored PTMs.[7f-h]

Another big emphasis of our group, which is highlighted in this presentation, is the generation of novel peptide- and protein-conjugates for pharmaceutical and medicinal applications as well as the cellular delivery of functional proteins and antibodies. For the latter study we employed cyclic cell penetrating peptides (cCPPs) to transport a functional full length protein to the cytosol of living cells as recently demonstrated by the direct delivery of GFP-conjugates.[8] For protein modification we use a combined approach of intein expression as well as recently developed bioorthogonal reactions and enzymatic ligations, for instance the so-called Tub-tag labeling.[9] This concept is finally applied to generate new antibody-drug conjugates as well as cell-permeable nanobodies, i.e. small antigen binding proteins that remain active within the reductive milieu inside living cells, to interfere with intracellular targets.[10]

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