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A Series of Vectors for the Production of In-Vivo Biotinylated Recombinant Proteins in Different Hosts

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Site-specific biotinylation is of great interest for directed immobilisation of recombinant proteins. We have designed a streamlined series of vectors for the in vivo biotinylation of recombinant proteins in a range of hosts such as *Escherichia coli*, mammalian cells and *Leishmania tarentolae*.

All vectors possess the same multi-cloning site and result in N-terminal fusion proteins with a hexahistidine- and a biotin acceptor-tag (AVI-tag). The AVI-tag is specifically recognised by the BirA biotin ligase and mono-biotinylated in vivo. To allow for efficient biotinylation, each host has been specifically modified to express BirA either by addition of specific helper plasmids, co-expression of the biotin ligase from the same vector or by stable integration of the gene into the host genome.

Here, we will show the design strategies employed and give some examples for the production of soluble in-vivo biotinylated proteins in each expression system.