

From: (Maiuri et al., 2016)

Huntingtin-specific Chromobodies:

The HCB1 vector was generated by PCR amplifying the Hap1 huntingtin-specific intrabody recognizing amino acids 41-81 of huntingtin (Southwell et al., 2008) with a four-glycine linker at its C-terminus, as well as Sall and BspEI sites. The amplicon was ligated into a pEYFPN1 vector bearing a pEYFPC1 multiple cloning site using the aforementioned sites. The HCB2 vector was generated using the Gibson Assembly approach. iVHH4 huntingtin-specific intrabody recognizing amino acids 49-148 of huntingtin (Schut et al., 2015) was amplified with homology regions corresponding to modified pEYFPN1 vector surrounding the BamHI site. To generate nucHCB1 and nucHCB2, one SV40 NLS was added to each construct at the C-terminus of EYFP using the Gibson Assembly method by taking advantage of the C-terminal XbaI site. All Gibson Assemblies were carried out according to the manufacturer's guidelines using the Infusion kit (Clontech).

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