Reprogramming lipid synthesis in Chinese Hamster Ovary (CHO) cells for enhanced recombinant protein production

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The endoplasmic reticulum (ER) plays a critical role in protein folding, protein secretion, calcium homeostasis, and lipid biosynthesis. Mammalian cells are often used for the production of recombinant biotherapeutic proteins where the secretory pathway machinery, including the ER, is essential to the correct folding, assembly and post-translation modifications required of the target protein. However, expression of recombinant proteins in high amounts in mammalian cells can result in ER stress, which can result in cellular responses and multiple stimuli from the ER that activate the unfolded protein response (UPR), slow protein synthesis and can negatively impact upon protein yields and quality. The maintenance of the ER and secretory pathway system requires a carefully coordination of lipid biosynthesis. Here we investigate approaches and strategies to design new hosts and cellular circuits to reprogramme the CHO cell ER with a view to either expanding its capacity and/or subsequent secretory vesicle system to improve cell growth, yields and quality of recombinant secreted proteins. Our hypothesis is that controlled manipulation of lipid biosynthesis will result in an enhancement of the efficiency of the CHO platform as a recombinant protein expression system. Here we report on the manipulation of the CHO lipid biosynthesis machinery by altering key components. We have transiently and stably over-expressed two proteins in particular reported to led to expansion of the ER in CHO cells. Stable cell pools have subsequently been cloned via limited dilution cloning to obtain clonal cell lines. Over-expression of the lipid biosynthesis proteins did not impact upon cell growth behaviour, however transient expression of two model recombinant proteins (EPO and Etanercept – a TNFR-Fc fusion protein) that are difficult to express in CHO cells was enhanced in CHO cells engineered to over-express the lipid biosynthesis proteins. Here we present implications for this and potential applications.

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