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# Prediction of stable and transient expression of recombinant proteins from CHO cells based upon translational reprogramming

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# PREDICTION OF STABLE AND TRANSIENT EXPRESSION OF RECOMBINANT PROTEINS FROM CHO CELLS BASED UPON TRANSLATIONAL REPROGRAMMING

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Translational reprogramming and mRNA translation efficiency influence global protein synthesis, cell proliferation and growth; important parameters in defining recombinant protein expression yields. Reprogramming generally results in a down-regulation of overall global protein synthesis. Polysome profiling is used to analyse mRNA translation via the distribution of ribosomes between monosomes and polysomes, and can be used to investigate translational reprogramming occurring within the cell culture. Here this approach has been applied to investigate the endogenous polysome profiles of host and recombinant Chinese Hamster Ovary (CHO) monoclonal antibody-producing cell lines, and how the profiles change across culture depending on the growth and protein requirements of the cell.

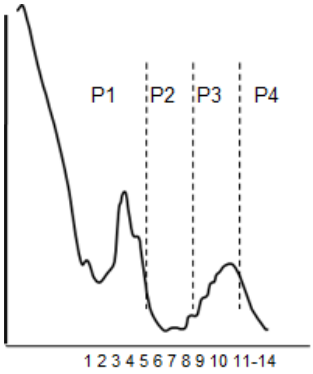


*Figure 1: Example CHO polysome profile showing the fractions collected and those pooled for qRT-PCR*

We report that polysome profiling (see Figure 1) reveals differences in the profiles of the 40S, 60S, 80S and polysome peaks between different stably expressing monoclonal antibody cell lines. Further, a direct comparison of transient and stable expression of the same target molecule defines how and when translational reprogramming occurs for the different expression systems. We also report how temperature-shift during culture influences translational reprogramming and that qRT-PCR analysis on mRNA isolated from the different fractions generated during polysome analysis to determine the copy number of recombinant mRNAs (GS, heavy chain, light chain) throughout culture reflects the number of mRNAs being actively translated and relates to product yield. We have also determined the amounts of particular mRNAs in the polysome fractions involved in the response to stresses that the cell may encounter during culture and recombinant protein production to identify how the cell reprograms to cope with such demands. These analyses show that as reprogramming occurs, there are changes in the distribution of particular mRNAs between cell lines and across culture. This data will allow us to understand mechanistically how the cell reprograms protein synthesis to cope with the demands of recombinant protein production.