ANALYSIS OF DNA DSB REPAIR AND PRODUCTION STABILITY IN CHO CELLS

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Productivity of recombinant proteins in CHO cell lines often decreases over long-term cultivation. This production instability limits the use of CHO-based platforms and can negatively impact the capability of a manufacturing process to meet market demands. A method to prevent the production loss during long-term cultivation is highly desirable. Genome instability can reduce transgene copy number and is reported as a major cause for production instability. We hypothesize that the DNA double-strand break (DSB) repair system in CHO is deficient and associated with both genome and production instabilities. Our results indicated that CHO cells had a lower DSB repair rate compared to the bEnd.3 mouse endothelial cell line, which is consistent with our hypothesis. The ability to improve DSB repair in CHO may provide a strategy to prevent production instability. Therefore, we tested heterologous expression of eight DSB repair-related genes, and found that four genes could significantly improve DSB repair in CHO cells. To further assess the impact of improved DSB repair on protein production, each of the four heterologous genes was stably expressed in a secreted alkaline phosphatase (SEAP) producing cell line, and SEAP production in single clones was evaluated over three months in the absence of methotrexate (MTX). Our results showed that productivity correlated strongly with the SEAP copy number, and two heterologous genes could substantially improve the production retention during long-term cultivation.