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Effective microRNAs for cell line engineering and cellular mechanisms of action

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MicroRNAs (miRNAs) constitute an important class of small non-coding RNAs and are imperative for the regulation of gene expression in mammalian cells. By post-transcriptionally modulating the expression of hundreds of different genes concomitantly, miRNAs are capable of regulating entire cellular pathways to control cellular phenotypes. Thus, miRNAs represent promising tools for cell engineering of biopharmaceutical production cells.
By taking advantage of a functional high-content miRNA screening procedure we were able to identify >750 miRNAs significantly affecting protein expression, cell growth, apoptosis and necrosis in CHO cells. Functionality of impactful miRNAs could be successfully validated in secondary screening approaches as well as by stable miRNA overexpression. Furthermore, we demonstrate that identified pro-productive miRNAs were able to substantially increase monoclonal antibody yields of two industrial high-producing CHO cell lines expressing different monoclonal antibodies. To better understand the molecular mechanisms behind this improved phenotype, we analyzed selected pro-productive miRNAs and were able to reveal downstream mRNA targets and underlying cellular mechanisms of action. In this conjunction, we identified the ubiquitin pathway to be involved in enhanced protein production induced by the miR-30 family, miR-2861 was discovered as a potential HDAC5 inhibitor enhancing productivity in CHO cells while maintaining product quality and miR-483 was shown to act as species independent universal enhancer of cellular antibody and viral productivity while mimicking mild hypothermia.
These significant findings reveal not only the existence of effective miRNAs for the use in cell line engineering but also shed light on underlying mechanisms of action within the cellular environment supporting the hypothesis that miRNAs regulate diverse and essential cellular pathways important for industrial protein production. Engineering production cell lines using miRNAs therefore constitutes a highly attractive and easy-to-use methodology to substantially enhance cellular productivity.