Enhanced Sialylation of Recombinant Erythropoietin in Chinese Hamster Ovary Cells by Expression of Sialyltransferase and Galactosyltransferas

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Abstract

Human 2,3-sialyltransferase (ST) and 1,4-galactosyltransferase (GT) were engineered into CHO cells which produce human EPO in order to enhance the sialylation of the protein. ST and GT are responsible for the terminal sialylation and galactosylation, respectively. Recombinant human EPO was purified from the culture supernatant of the engineered CHO cells by an immuno-affinity chromatography. N-Glycans were isolated after digestion with glycoamidase-F, and the sialylation of EPO was evaluated by HPLC. When the 2,3-ST was expressed in CHO cells (EC1-ST2), the sialylation of EPO glycans was significantly increased compared with that of control cells (EC1). In detail, the amounts of di-, tri-, and tetra- sialylated glycans were slightly increased while those of asialo- (neutral) and mono-sialylated glycans were decreased. Specially, the amount of tri-sialylated glycans was increased from 17.3 % to 26.1 %. When both 2,3-ST and 1,4-GT were coexpressed in CHO cells (EC1-GTST15), the sialylation was more increased than that of EC1-ST2. In case of EC1-GTST15 cells, the portion of tri-sialylated glycans was remarkably increased from 17.3% to 35.5%. The expressions of ST and GT in CHO cells did not affect both cell physiology and culture environment including cell growth, EPO productivity, glucose consumption rate, lactate production rate, and culture pH. Thus, coexpression of 2,3-ST and 1,4-GT might be beneficial in CHO cells for producing a therapeutic glycoprotein with enhanced sialylation.

References

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