E109 Characterization of the human folate receptor (hFR)-γ produced by CHO cells.

Won-Sin Kim
Division of Life Science, Wonkwang University

The human folate receptor (hFR) is a plasma membrane protein that is anchored to the membrane via a glycosylphosphatidylinosito (GPI) tail in some cell types. However, the hFR-γ has a truncated hydrophobic, α-helical carboxyl terminus that is thought to be the signal for cleavage and attachment of the GPI tail. Thus, hFR-γ was suggested as a secretory type protein due to lack of an efficient signal for GPI modification. In this study, we transfected the hFR-γ cDNA into Chinese hamster ovary (CHO) to investigate its biosynthesis and binding affinity. The hFR-γ-expressing CHO cells produced a [^{3}H] folic acid binding protein in the medium. However, we couldn't detect any cell surface [^{3}H] folic acid binding and transport activities. The hFR-γ-expressing CHO cells were more rapidly damaged by folic acid depletion than normal CHO and hFR-α-expression cells in the low concentration folic acid media (DMEM without added folic acid, 1% FCS). Result of these experiment suggest that although soluble type hFR-γ produced by CHO cells can bind [^{3}H] folic acid, they are not involve in transport of folic acid across plasma membrane.

E110 **Induction of p53-independent p21 during ceramide-mediated G1 arrest in Human Hepatoma cells.**

Kyung-Hwa Kang, Won-Ho Kim, Do-Hyun Chae, [1] Mie-Young Kim and Kyung-Hee Choi.
Department of Biology, College of Natural Sciences, [1]College of Pharmacy, Chung-Ang University, Seoul 156-756

Effects of ceramide on the induction of G1 arrest and the involvement of p53-independent p21 in G1 arrest were studied in human hepatoma cell line, SK-Hep1 cells. Ceramide-induced G1 arrest was detected after 6 hours and 80 % of the cells were accumulated in G1 phase after 24 hours. Dephosphorylation of pRb was induced by ceramide and increased as cells were arrested in G1 phase. The level of p21 mRNA and protein were increased by 6 hours after exposure to ceramide, when pRb dephosphorylation was initiated, and increased with longer treatment. However, the expression level of p53 was not changed. Then, we tested whether p53-independent p21 protein induced by ceramide is involved in inhibiting activity of Cdks. The kinase activity of cyclin E/Cdk2 protein was decreased in response to ceramide. Furthermore, the association of p21 with cyclin E/Cdk2 complex was increased compared with that of control. These results suggest that ceramide induces p21 through p53-independent manner, which is responsible for the inhibition of cyclin E/Cdk2 and pRb dephosphorylation, and it is at least one of the mechanism to mediate ceramide-induced G1 arrest.