

Z605 Caspase-mediated Cleavage of p65 (Rel A) during VK₃ Derivative, C11-induced Apoptosis

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Nuclear factor (NF)- κ B is implicated to protect cells from apoptosis induced by TNF and various genotoxic agents. In this study, we demonstrated that addition of C11, a VK₃ derivative resulted in repression of NF- κ B activity by inducing the proteolytic cleavage of NF- κ B p65 (Rel A) and apoptosis in HeLa cells. Cleavage of p65 (Rel A) was occurred earlier than the cleavage of PARP and DNA fragmentation during C11-induced apoptosis. Pretreatment of TNF α suppressed the C11-induced cleavage of p65 (Rel A). In vitro, p65 (Rel A) was cleaved by caspase 3 at N-terminal region rather than C-terminal region and its cleavage was prevented by Ac-YVAD-CHO and Ac-ZVAD-CHO, inhibitors of caspases. Caspase 1 and 8 had no effect upon the cleavage, and p50, another subunit of NF- κ B, was not cleaved by C11. These results suggest that NF- κ B inactivation through p65 (Rel A) N-terminal cleavage by caspase 3 is partially involved in C11-induced apoptosis of HeLa cells.

Z606 Effect of Inhibitors on DNA-Protein Crosslink Formation in Cross-adapted CHO Cells

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The present study was investigated the effect of inhibitors on the DNA-protein crosslink (DPC) to ethyl methanesulfonate (EMS) or ultraviolet C (UVC) in cross-adapted Chinese hamster ovary cells. To investigate the effect of inhibitors on the induction of DPC to EMS or UVC, we used K⁺-SDS (potassium ion-sodium dodecyl sulfate) precipitation method, and 2,4-dinitrophenol (DNP), reducing agents of intracellular ATP, 2',3'-dideoxythymidine 5'-triphosphate (ddTTP), an inhibitor of polymerase β , 3-aminobenzamide (3AB), an inhibitor of poly(ADP-ribosyl)ation, novobiocin (NOV), an inhibitor of type II eukaryotic DNA topoisomerase and cyclohexamide (CHM), an inhibitor of protein synthesis. The treatment with 50 μ M DNP during 4 hour incubation after 2 mM EMS decreased the amount of DPC induced by 5 J/m² UVC. The treatment with 1 μ M CHM after 4 hour incubation after 2 mM EMS or 1 J/m² UVC increased the amount of DPC induced by 5 J/m² UVC or 8 mM EMS.