INDUCTION OF APOPTOSIS BY RESVERATROL IN CANCER CELLS DURING COMBINATION TREATMENT WITH NUTLIN-3 AND TGF-β

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INTRODUCTION. Resveratrol (3, 4’, 5-trihydroxy-trans-stilbene) is a naturally occurring compound found mainly in the skins of grapes and various other fruits and nuts. Resveratrol (RV) has been previously studied and found to work through various mechanisms and produce anti-inflammatory, antiviral and anticancer effects (1,2,3). Previous studies have shown that RV promotes p53-dependent apoptosis via a Ras-MAPK signal transduction pathway by increasing the levels of serine phosphorylation of p53 (4). Alteration of expression of Bcl2 family proteins, loss of mitochondrial function, release of cytochrome c, and activation of caspases are also believed to be involved in the RV induced cell death (5).

METHOD. Our aim for this study was to investigate the induction of apoptosis by RV (5 µM) in A2780/CP-70 cells in combination with Nutlin-3 and TGF-β. The combination treatments with Nutlin-3 (10 µM) and TGF-β (1 µg/ml) were used to enhance RV induced apoptosis through synergistic mechanisms. The cells were incubated with RV in the presence or absence of Nutlin-3 or TGF-β for 24 and 48 hrs. At the end of incubation periods the cells were probed using DAPI (4’,6-diamidino-2-phenylindole) for the detection of apoptosis (see Fig. 1 and Fig. 2). The extent of apoptosis was also confirmed by analyzing the DNA ladder formation.
Fig. 1. Representative picture of control cells probed with DAPI after a 24 hr. incubation period.

Fig. 2. Representative picture of RV + Nutlin-3 treated cells probed with DAPI after a 24 hr. incubation period.

DISCUSSION. Resveratrol (RV) has been shown to produce apoptotic effects in cancer cells around 25 µM concentrations. In our experiments, the treatment of A2780/CP-70 cells with lower concentrations (5 µM) of RV alone did not show much apoptosis in 24 - 48 hrs. However, the combination treatment with Nutlin-3 or TGF-β showed apoptosis starting from 24 hrs of treatment. The concentration of RV used in mono-therapy is at least 4 times lower than the concentration typically used for effectively inducing apoptosis in cancer cells. It appears that combining lower concentrations of RV with Nutlin-3 or TGF-β is equally effective as high concentrations of RV in cancer cells for inducing apoptosis.

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REFERENCES.