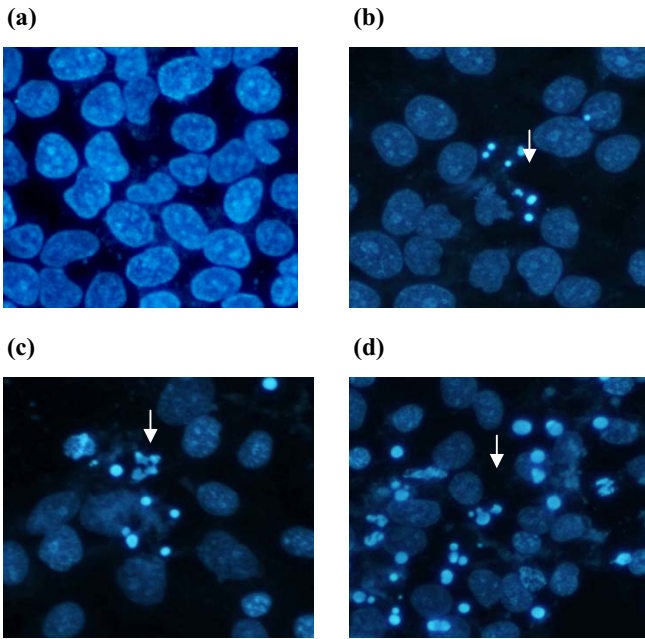
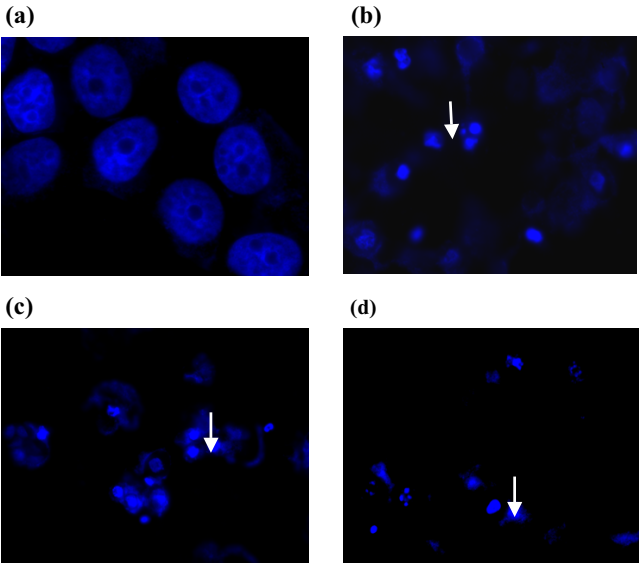


Figure 5



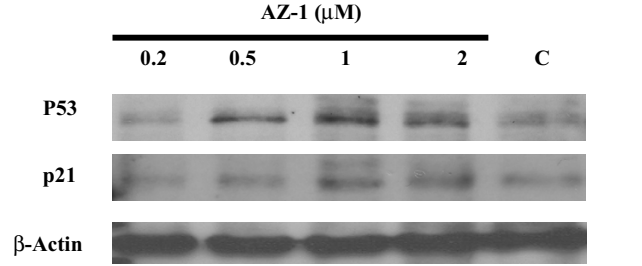
Apoptosis induced by bis-aziridinylnaphthoquinone (AZ-1) in BC-M1 cells. The BC-M1 cells were treated in 24-hour culture with (a) dimethylsulfoxide control, (b) 0.2 μ M AZ-1, (c) 1.0 μ M AZ-1, or (d) 2.0 μ M AZ-1 before Hoechst staining and analysis of the DNA nuclei. Apoptotic cells showed as blue, peripherally clumped or fragmented chromatin as indicated by the arrows.

Figure 6



Apoptosis induced by bis-aziridinylnaphthoquinone (AZ-1) in MCF-7 cells. The MCF-7 cells were treated in 24-hour culture with (a) dimethylsulfoxide control, (b) 0.2 μ M AZ-1, (c) 1.0 μ M AZ-1, or (d) 2.0 μ M AZ-1 before Hoechst staining and analysis of the DNA nuclei. Apoptotic cells showed as blue, peripherally clumped or fragmented chromatin as indicated by the arrows.

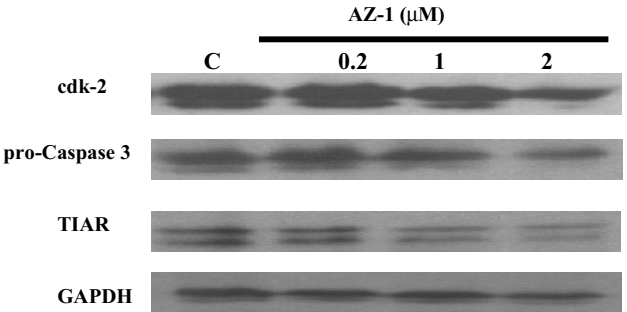
Figure 7



| Protein | AZ-1 (μ M) | | | | C |
|---------|-----------------|------|------|------|---|
| | 0.2 | 0.5 | 1 | 2 | |
| p53 | 0.96 | 1.11 | 1.43 | 1.31 | 1 |
| p21 | 1.03 | 1.06 | 1.18 | 1.22 | 1 |

Bis-aziridinylnaphthoquinone (AZ-1) alters the expression of apoptotic proteins p53 and p21. The protein expression of p53 and p21 were assessed by immunoblot. The BC-M1 cells were treated with various concentration of AZ-1 (lanes 1–4: 0.2 μ M, 0.5 μ M, 1.0 μ M, and 2.0 μ M) for 24 hours and cell lysate was prepared for analysis. Lane 5, untreated control (C). β -actin was an internal control on BC-M1 cells.

Figure 8



| Protein | C | AZ-1 (μ M) | | |
|-------------|---|-----------------|------|------|
| | | 0.2 | 1 | 2 |
| TIAR | 1 | 1.02 | 0.62 | 0.40 |
| pro-Caspase | 1 | 1.19 | 0.85 | 0.67 |
| cdk-2 | 1 | 1.07 | 0.65 | 0.57 |

Bis-aziridinylnaphthoquinone (AZ-1) alters the expression of apoptotic proteins. The protein expression of T-cell restricted intracellular antigen-related protein (TIAR), pro-caspase 3 and cyclin-dependent kinase (cdk2) were assessed by immunoblot. The BC-M1 cells were treated with various concentration of AZ-1 (lanes 2–4: 0.2 μ M, 1.0 μ M, and 2.0 μ M) for 24 hours and cell lysate was prepared for analysis. Lane 1, untreated control (C). GAPDH was an internal control on BC-M1 cells.