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Mechanism study on DNA damage and Apoptosis induced by heak shock using Comet Assay

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Comet assay, single cell gel electrophoresis has been known as useful, rapid, simple, visual, and sensitive technique for measuring the DNA breakage in mammalian cells. For evaluation of DNA damage using comet assay, early studies reported a change in comet length and intensity with DNA damage using simple visual technique, such as fluorescence microscopy with eyespiece. In recent, some workers are observing and analyzing nucleotide of comets using quantitative fluorescence image analysis system to estimate 'tail moment', which is defined as the product of the tail length and the fraction of total DNA in tail. Our laboratory also adopted the image analysis software for quantification.

In addition, many of the practical features of comet assay render it potentially attractive as useful tool for molecular toxicology and carcinogenesis, because the system is already showing considerable promise as rapid predictor in both *in vitro* and *in vivo* experimental designs. Recently, the comet assay becomes a attractive technique to study of apoptosis, because apoptotic fragmentation of nuclear DNA into nucleosomal sizes can be evaluated by the comet assay. So, we attempted to apply the comet assay to studying the effect of various stress on the apoptosis-sensitive cell lines. Particularly, focusing on the hyperthermic apoptosis, we could find that heat shock(44°C for 60 minutes) was sufficient to induced apoptosis in these cell lines. But using the highly sensitive comet assay, we could not detect DNA breaks immediately after heat shock.

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