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# Use of the comet assay technique for quick and reliable prediction of *in vitro* response to chemotherapeutics in breast and colon cancer

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## Abstract

**Background:** Determination of response to chemotherapy is a major requirement of personalized medicine. Resistance, whether developed or native, critically affects a treatment's success. Single Cell Gel Electrophoresis - also known as a comet assay - is used to detect DNA damage at the level of individual eukaryotic cells. We assessed the use of comet assays in determining response to chemotherapeutic drugs that are widely used in breast and colon cancer.

**Results:** We treated human breast and colon cancer cell lines with melphalan, cisplatin, mechlorethamine or doxorubicin, as monotherapies. Drug activities varied even in the same cancer types, further demonstrating the heterogeneity of different cancer types.

**Conclusion:** The comet assay technique can provide reliable and quick results with minimum requirements and is applicable to a wide variety of drugs.

**Keywords:** Comet assay, Personalized medicine, Breast cancer, Colon cancer, Cisplatin, Melphalan, Mechlorethamine, Doxorubicin

## Background

Personalized medicine requires that therapy should be customized to individual patients, using genetic or other information [1]. Prediction of response to chemotherapy drugs is a major concern in cancer treatment [2]. Resistance to chemotherapy agents may exist before, or develop during, therapy [3]. Most techniques to predict response entail analysis of expression of different genes [4]. However, fast, reliable results are urgently needed; moreover, a drug's effect cannot always be predicted by measuring gene expression [5].

Single Cell Gel Electrophoresis (SCGE), also known as comet assay can measure DNA damage in individual eukaryotic cells. The principle of the comet assay is that unfragmented DNA maintains a well-organized structure in the nucleus, but becomes disrupted when the cell is damaged. It detects both single-strand and double-strand breaks, and has a simple and inexpensive setup. Comet assay is therefore a promising technique for

predicting response to drugs that are affected by DNA structure [6].

The present study evaluated the predictability of response to widely used chemotherapy drug from comet assay results in established human breast and colon cancer cell lines. We tested nitrogen-mustard alkylating agents (melphalan, mechlorethamine) and doxorubicin in breast cancer, and tested cisplatin in colon cancer cell lines.

## Results and discussion

Analysis of results was based on percentages of DNA in the comet "head" (amount of genetic material distributed in the nucleus) and in "tail" (amount of genetic material distributed in the fragmented pieces). We examined  $\geq 100$  cells for each combination of cells and drugs. With  $p$  set to be 0.05, we estimated the ranges of DNA percentage in untreated and treated cells; where these ranges overlapped, the cells were rejected. In the MCF-7 and MDA-MB 231 cell lines, we clearly observed functional activity for all drugs. The T47D cells showed

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