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Final Report on *A retroelement based diagnostic assay for changes in chromatin status during cancer development*

John F. McDonald, PI

In the study that was funded by OCI, we proposed to develop and test the feasibility of using a human retroelement microarray as a marker of epigenetic changes associated with cancer development. Four specific projects were outlined. As documented below, we have successfully completed projects 1 & 2. Project 3 is currently in progress. Project 4 will be completed over the second half of the funding.

Project 1. Design of retroelement probes specific for > 650 families of human retroelements (LINEs, SINEs, HERVs, etc,);

Results:
We have completed the design of 670 retroelement probes representing distinct families of human retroelements. We have filed two patents on the use of these probes in a microarray bioassay of changes in methylation patterns in early staged tumors (Global analysis of transposable elements as molecular markers of cancer: Provisional Patent # 60/466,798) and in embryonic stem cells (Global analysis of transposable elements as molecular markers of the developmental potential of stem cells: Provisional Patent # 60/466).

Project 2. Select a subset of representative oligonucleotides probes to produce a prototype spotted retroelement array;

Results:

We have developed the prototype retroelement array and have demonstrated that it can detect variation in retroelement expression among ovarian malignant and non-malignant tissue samples.

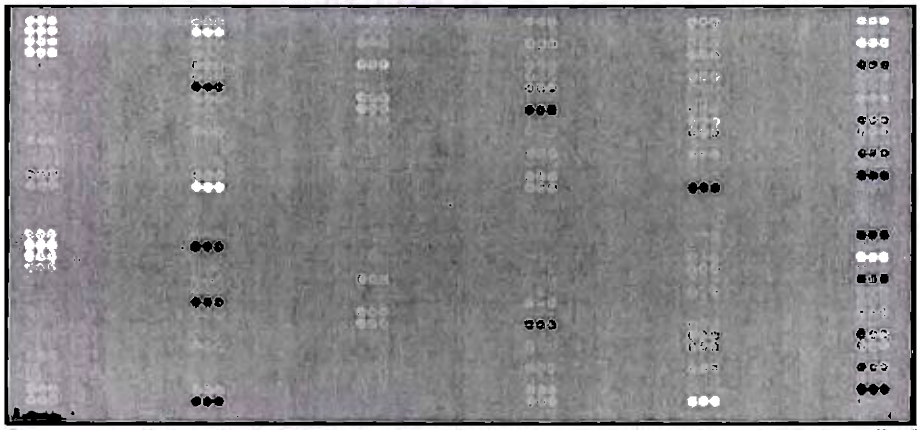

Figure 1. Prototype spotted retroelement array hybridized against RNA isolated from the HeLa cell line and a breast cancer cell line (HS 578t). Three replicate oligos are spotted horizontally. This prototype array is being used to monitor expression and methylation patterns of retroelements (and other transposable elements) in a various stages and types of ovarian cancer.

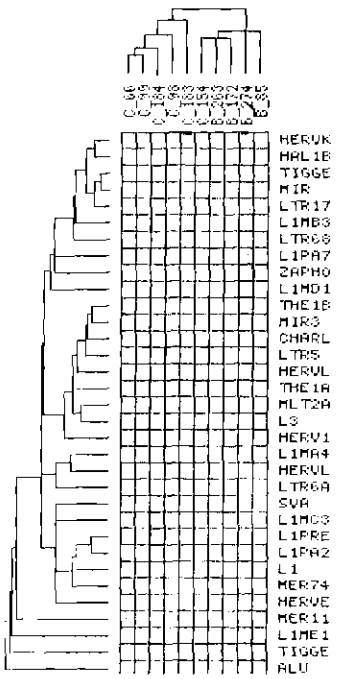

Figure 2. Results from preliminary analysis of retreolement expression in ovarian cancer (Stage III, adenocarcinomas-serous) and benign (adenomas) tissue samples. Shown is a 2-way hierarchical clustering of microarray expression data from our prototype retroelement sarray. The preliminary results indicate that differences in expression of the some retroelement may be a good predictor of ovarian cancer.

Project 3. Utilize the prototype array to monitor retroelement expression in a series of ovarian carcinomas, non-malignant ovarian adenomas and normal ovarian tissue.

Result

We have demonstrated that retroelement expression profiles can reliably distinguish between ovarian tumors (Figure 2).

Project 4. Utilize the prototype array to test if differences in retroelement expression are correlated with differences in the methylation status of retroelements in the above malignant and non-malignant ovarian tissues.

Result:

We were unable to complete this aspect of the study due to the fact that we ran out of funds. However, based upon the preliminary results described above, we have been able to apply for and receive grant from the Ovarian Cancer Fund, Inc. to continue these studies.

Concluding Comment.

This grant from the Georgia Cancer Coalition enabled us to acquire valuable preliminary results that have served as the basis of two grant proposal recently submitted to the Federal Government for funding. One to the Department of Defense Ovarian Cancer Program for $298,721 and another to National Institutes of Health ($350,000).