

Project Title: Dedicated Flow Cytometer for Monitoring of Body Cavity Fluids in Lung Cancer Patients

Project Summary: Lung cancer screening is primarily based on radiographic examination (chest X-rays) of the patients. Cytopathology of bronchial brushing and washing, bronchi alveolar lavage and of pleural fluid samples has been used for diagnostic purpose. As cytopathology is based on morphological examination of a small number of cells under a microscope, approximately one half of the body cavity fluid samples with malignant cells are not correctly diagnosed, thus resulting in a 50% rate of false negatives. Thus there is an urgent need for improving detection of tumor cells in body cavity fluids by incorporating rapid analytical methods such as immunocytochemistry in combination with high-resolution flow cytometry. Using these highly quantitative and specific methodologies, a large number of cells in body cavity fluids can be rapidly screened and cellular markers of malignancy and cellular origin identified.

Although flow cytometry is extensively used for phenotypic analysis of leukemia, its use as a diagnostic tool for analysis of pleural fluids has been limited. Some of the reasons for relative lack of flow cytometric studies on body fluids are related to the large number of false positives reported in earlier studies based on detection of DNA aneuploidy. The second main reason is related to the complexity of the modern commercially available flow cytometers, which are often equipped with several lasers and sophisticated optical systems for multiparametric studies. These refinements are reflected in the overall complexity and cost of the instruments (which can often exceed more than \$ 200,000) and require expensive service contracts and supervision by a highly trained flow cytometer operator.

Our hypothesis is that a low cost high resolution flow cytometer which rapidly measures nuclear volume versus DNA content along with the expression of specific markers can be developed for rapid screening of cells in body cavity fluids of patients suspected to have a malignancy. Specifically in patients suspected to have a smoking related lung malignancy, this instrument could screen bronchial washings and pleural fluid samples to identify cells with aneuploid DNA content and for the expression of markers such as TTF, which is specific to cells of pulmonary origin. To be useful, this instrument will have to be low cost, require very little technical expertise for operation and capable of auto-analyzing the data. We propose to work with NPE Systems of Pembroke Pines, FL to develop and test such a flow cytometer for screening of body cavity fluids. By correlating data obtained from this high resolution, low cost dedicated flow cytometer with conventional diagnostic cytopathology, we may be able to improve detection of tumor cells in body cavity fluids of lung cancer patients.