![MTTI logo]

Molecular Targeting Technologies, Inc.

Development Pipeline

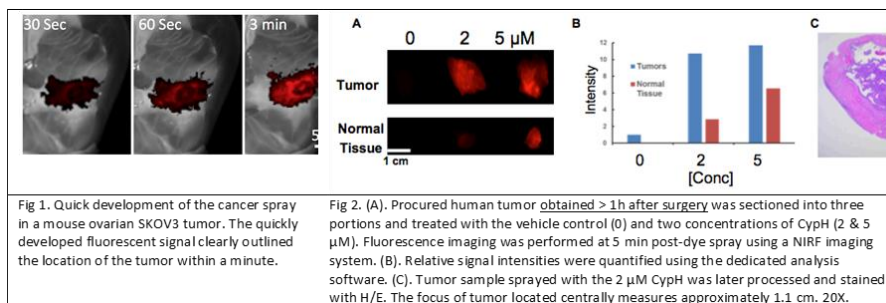**CLASS:** Diagnostic Spray for Guided Surgery

NAME: CypH

INDICATION: Ovarian cancer surgery

USES: To augment the visual detection of small tumors and tumor margins for accurate biopsy, complete resection, and enhanced second-look surveillance during already scheduled ovarian cancer surgical procedures.

TECHNOLOGY: Most cancers show a dysregulated metabolism and resulting acid-base changes in the tumor microenvironment. in cancer cells the intracellular pH and the extracellular pH are ≥ 7.4 and ~6.4 - 7.1, vs. 7.2 and 7.4 in normal cells. We target the tumor extracellular acidic environment with a fluorogenic dye (CypH). It is non-fluorescent in normal tissues but generates a high-contrast fluorescence signal for visible detection of malignant tumors. CypH activates within minutes of application during surgery. A small amount of CypH is sprayed into the patient to see remaining cancerous tissue. Other probes require large IV doses hours or days before surgery, raising the risks of toxic or allergic reactions.

UNMET NEED: Epithelial ovarian cancer is the most lethal gynecologic malignancy. Most women are diagnosed late due to few symptoms in early disease and no effective screening methods. Treatment usually includes surgery followed by chemotherapy. Post-chemotherapy, most women are declared to be in clinical complete response. During surgery, surgeons cannot see where tumor tissue ends and healthy tissue begins. Residual disease, invisible cancerous tissue "seeds" which metastasize, is usually below the level of detection by physical examination, tumor markers, radiologic evaluation and second-look white light laparoscopy. >80 of women experience recurrence. An imaging platform that can reliably locate these subclinical lesions is urgently needed to improve surgical outcomes.

PROOF OF CONCEPT: An ovarian SKOV3 tumor was subcutaneously inoculated in mice. When tumors were 2-3 mm, the skin was removed and the CypH solution (2µM) was sprayed onto the tumors. No fluorescent signal was observed immediately after the spray. A fluorescent signal outlining the tumor quickly developed within a minute of the spray's application (Fig 1). The contrast continued to increase and quickly reached plateau at ~ 5 min. Using freshly harvested human ovarian tumor tissue and adjacent normal tissue, two concentrations of CypH and a vehicle control were sprayed onto all tissue samples and then imaged with a CCD camera at a total elapsed time of 5 minutes after dye application (Fig. 2A). Semi-quantitative analysis of the fluorescence intensity reveals ~3.8-fold greater signal in the tumor relative to normal tissue at the 2µM CypH concentration (Fig. 2B). Histochemical analysis showed good agreement with the fluorescence image (Fig 2C shows a representative photomicrograph). These experiments suggest that a pH dependent cyanine dye could be used as an aerosol spray for near real-time tumor detection.

Fig 1. Quick development of the cancer spray in a mouse ovarian SKOV3 tumor. The quickly developed fluorescent signal clearly outlined the location of the tumor within a minute.

Fig 2. (A). Procured human tumor obtained > 1h after surgery was sectioned into three portions and treated with the vehicle control (0) and two concentrations of CypH (2 & 5 µM). Fluorescence imaging was performed at 5 min post-dye spray using a NIRF imaging system. (B). Relative signal intensities were quantified using the dedicated analysis software. (C). Tumor sample sprayed with the 2 µM CypH was later processed and stained with H/E. The focus of tumor located centrally measures approximately 1.1 cm. 20X.

STAGE OF DEVELOPMENT: Preclinical. Seeking partner.

PRINCIPAL COLLABORATOR: Cornell Medical College

IP: Pending patents owned by Methodist Hospital, Houston, Texas

FUNDING: Secured funding from NCI

OWNERSHIP: MTTI is establishing an option agreement with Methodist Hospital