

Manual quantitation of B16F10 pulmonary tumor colonies in mice correlates with ^{18}F FLT uptake.

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Introduction

The C57Bl/6 syngeneic murine model of melanoma metastasis using B16F10 tumor cells was established more than 30 years ago. Intravenous injection of B16F10 melanoma cells results in the formation of lung colonies that can be seen at 15 days post injection. Due to the pigmented nature of the tumor cells, the most common technique for analyzing tumor growth within the murine lung requires the post-mortem extraction of the lung block followed by a visual assessment and "quantification" of black tumor foci on the lung surface. In addition the tumor burden can be estimated using a qualitative scoring system (0 – 4+) that takes into account both number and size of tumors. Uptake of the cell proliferation tracer, 3'deoxy-3'-[^{18}F]fluorothymidine (^{18}F FLT), has been shown to correlate with cellular markers of proliferation in human, non small cell lung cancer, and because it is not sequestered by the myocardium, it provides better images of lung tumors than does ^{18}F FDG.

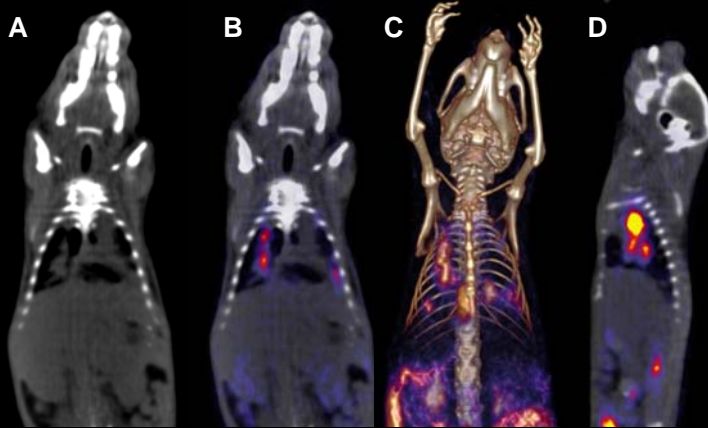


Figure 1. PET/CT imaging of pulmonary colonies of B16F10 melanoma in mice using ^{18}F FLT. Coronal plane pulmonary tumors revealed in CT (A) and fused PET/CT (B) images. (C) Three dimensional representation of ^{18}F FLT uptake in a mouse with pulmonary B16F10 tumors. (D) Sagittal view of PET/CT imaging of lung tumors with ^{18}F FLT

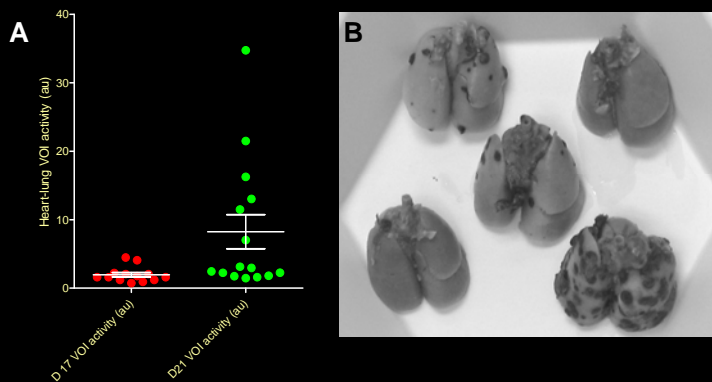


Figure 2. Pulmonary tumor burden appears at 17 d post injection of cells and progresses to day 21. (A) The ^{18}F FLT radioactivity associated with the heart-lung block volume of interest (VOI) increases significantly from day 17 to day 21 post cell injection. (B) At day 21 the lungs contain visible but variable amounts of tumor visible on their surface.

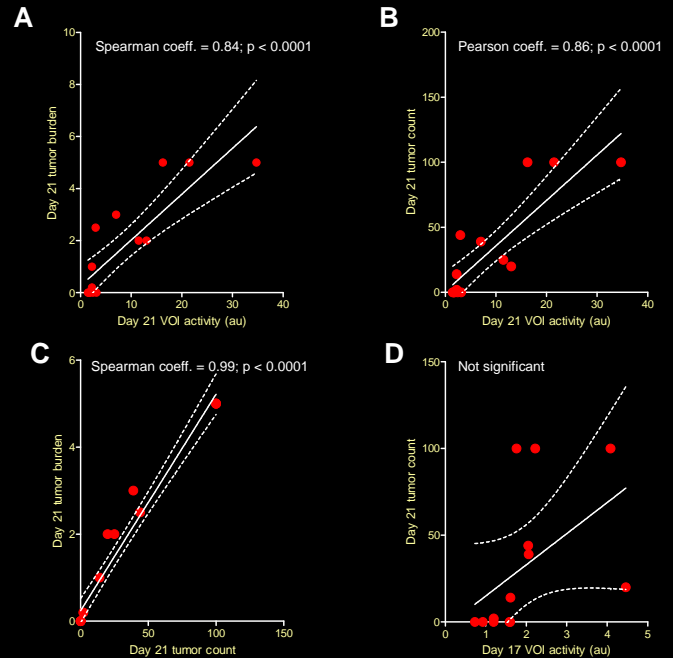


Figure 3. Correlation between invasive and non-invasive (^{18}F FLT imaging) methods for determining B16F10 pulmonary tumor load. Significant correlation between ^{18}F FLT retention in the whole heart-lung VOI and day 21 tumor burden (0 - 5+; surface scoring (A)) and tumor count (B) was observed. As expected, there was a highly significant correlation between the invasive methods of tumor burden and tumor counting (D). There was no correlation between ^{18}F FLT retention at day 17 and tumor count at day 21.

Results and Discussion

In this study, we compare the efficacy of estimating lung tumor burden in mice with ^{18}F FLT by using small animal PET/CT imaging with surface tumor counting and qualitative scoring. C57Bl/6 mice ($n = 15$) received 7.5×10^4 B16F10 melanoma cells via the lateral tail vein and tumors were allowed to develop for 17 days. At this time, the mice were anesthetized and PET/CT images acquired 1 h after iv injection of ~ 7.4 MBq of ^{18}F FLT. After a further 4 d of tumor growth, the mice were injected, euthanized at 1 h, imaged as before using ^{18}F FLT and the heart lung blocks excised at necropsy. The number of tumors visible on the surface of the 5 lobes was determined, and the tumor load was scored from 0 – 4+. Radioactivity (Bq/cc) contained within a manually segmented 3D volume of interest encompassing the heart and lung (but not the thymus) was measured, and a correlation analysis was performed.

^{18}F FLT was readily detected in pulmonary B16F10 colonies at both 17 and 21 d post-injection of cells. As expected, the day 21 ex vivo tumor count and tumor burden correlated significantly (Spearman coeff. = 0.99; $p < 0.0001$). Radioactivity within the lung volume of interest (VOI) at day 21 post injection correlated significantly with tumor colony counts (Pearson coeff. = 0.86; $p < 0.0001$) and estimated total lung tumor burden (Spearman coeff. = 0.84; $p < 0.0001$). The day 17 VOI activity correlated with day 21 tumor burden (Spearman coeff. = 0.82; $p < 0.001$) but did not achieve significance with the day 21 tumor count. These data support the use of ^{18}F FLT PET imaging to non-invasively and quantitatively evaluate the effect of various therapies on the growth of B16F10 lung colonies.