Identification and monitoring of sub-tumor targets will be a critical step for optimal design and evaluation of cancer therapies in general and biologically targeted radiotherapy (dose-painting) in particular. Quantitative PET imaging may be an important tool for these applications. Currently radiotherapy planning accounts for tumor motion by applying geometric margins. These margins create a motion envelope to encompass the most probable positions of the tumor, while also maintaining the appropriate tumor control and normal tissue complication probabilities. This motion envelope is effective for uniform dose prescriptions where the therapeutic dose is conformed to the external margins of the tumor. However, much research is needed to establish the equivalent margins for non-uniform fields, where multiple biological targets are present and each target is prescribed its own dose level. Additionally, the size of the biological targets and close proximity make it impractical to apply planning margins on the sub-tumor level. Also, the extent of high dose regions must be limited to avoid excessive dose to the surrounding tissue. As such, this research project is an investigation of the uncertainty within quantitative PET images of moving and displaced dose-painting targets, and an investigation of the residual errors that remain after motion management. This included characterization of the changes in PET voxel-values as objects are moved relative to the discrete sampling interval of PET imaging systems (SPECIFIC AIM 1). Additionally, the repeatability of PET distributions and the delineating dose-painting targets were measured (SPECIFIC AIM 2). The effect of imaging uncertainty on the dose distributions designed using these images (SPECIFIC AIM 3) has also been investigated. This project also included analysis of methods to minimize motion during PET imaging and reduce the dosimetric impact of motion/position-induced imaging uncertainty (SPECIFIC AIM 4).