A Revised Method for Inducing Secondary Lymphedema in the Hindlimb of Mice

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Background and Aim: Animal models are of paramount importance in the research of lymphedema in order to understand the pathophysiology of the disease but also to explore potential treatment options. This protocol will hopefully enable researchers to consider the limitations and advantages of the revised lymphedema model. The protocol should also assist researchers to successfully replicate the model.

Material and Method: The model requires that the mice get a dose of 10 Gray (Gy) radiation before and after surgery. The surgical part of the model involves ligation of three lymph vessels and extraction of two lymph nodes from the mouse hindlimb. Having access to microsurgical tools and a microscope is essential, due to the small anatomical structures of mice.

Results: The procedure has previously been used in three separate experiments. All the experiments were made by different lead investigators who all are co-authors of the article. Volumes of the hindlimbs were the primary outcome in all three experiments. We found that the mouse model allows researchers to induce significant lymphedema lasting at least 8 weeks.

Discussion/Conclusion: This method offers statistically significant lymphedema lasting at least 8 weeks, which has been measured directly via CT volumetric in three separate experiments by different lead investigators.

Quantification of tissue volume in the hindlimb of mice using microcomputed tomography images and analysing software

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Background and Aim: When studying illnesses that cause disturbance in volume such as lymphedema, reliable quantification of tissue volume is important. Experiments in rodent models provide a cost-effective research platform and are important for preclinical research on lymphedema. When performing such research, it can be crucial to measure the changes in tissue volume. Researchers must ensure that the risk of measurement error, when measuring the tissue volume, is as low as possible. The main goal of this article was to perform a comprehensive examination of the intra- and interrater agreement and hereby assess the risk of measurement error when using microcomputed tomography (µCT) images to measure hindlimb volume.

Material and Method: Four raters assessed scans of 50 mice once. Two of the four raters assessed the same scans twice. We then assessed the risk of measurement bias by examining the intra-and interrater agreement when using µCT-scans.

Results: The results of this study show that this technique has a low risk of measuring bias. The maximum estimated mean difference between pairs of raters was 0.26 mm^3 which is 0.17% of the mean hindlimb volume (150mm^3).

Discussion/Conclusion: This study might help researchers decide whether to use µCT for measuring hindlimb volume in mice. While being expensive and time-consuming, especially for raters new to the technique, the method has a very low risk of measuring bias and measures the volume of the hindlimb directly.