# Assessment of Accuracy and Reproducibility, as shown by Coefficient of Variation, using the iNSiGHT DXA System in both a Mouse and Rat Model

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## Introduction

The iNSiGHT Dual Energy X-Ray Absorptiometry (DXA/DEXA) system is a fully shielded DXA instrument used to measure bone mineral density (BMD) and bone mineral content (BMC), as well as lean and fat mass. It is a fully shielded system that can be integrated into any lab as there are no special infrastructure requirements to site the system. The purpose of this study, in collaboration with Dr. Val Fajardo at Brock University, was to assess the accuracy of the instrument in measuring BMD ($g/cm^2$), BMC (g), bone area ($cm^2$), tissue area ($cm^2$), fat percentage, fat mass (g), lean mass percentage, lean mass (g) and total weight (g) of the animal. The acquired data was then used to calculate a coefficient of variation (CV); results show a CV of less than 3% in all cases, highlighting the accuracy of the iNSiGHT system. Data was collected on the same animal multiple times, but also with the same animal re-positioned between each data set; again, the CV remained below 3%. This level of accuracy provides confidence in the reproducibility of the data collected using this system, and reduced inter-operator variability.

The iNSiGHT system is manufactured by Osteosys (Korea), and is distributed within North America by Scintica.

## Methods

All data was acquired using the iNSiGHT DXA system (Osteosys, Korea) in the lab of Dr. Val Fajardo at Brock University (St. Catharines, ON, Canada). All studies were conducted in accordance with approved animal protocols.

In these studies, male mice or rats were anesthetized using 2-3% isofluorane for the duration of the data collection; the acquisition time for each measurement was 25 seconds.

The animal was either repositioned by lifting them and placing them back onto the scanning bed or left untouched between each measurement, and in each case, BMD ($g/cm^2$), BMC (g), bone area ($cm^2$), tissue area ($cm^2$), fat percentage, fat mass (g), lean mass percentage, lean mass (g) and total weight (g) was measured at least 6 times. Mean, standard deviation, std error of mean and CV were then calculated using Microsoft excel. Data was then graphed using Prism for clarity.

## Results

The male mouse was either non-repositioned or repositioned and the DXA measurements were acquired 6 times (Figure 1). CV values in all cases were equal to or below 3%. SEM values were calculated (Figure 1, A and C) and were minimal for all measurements.

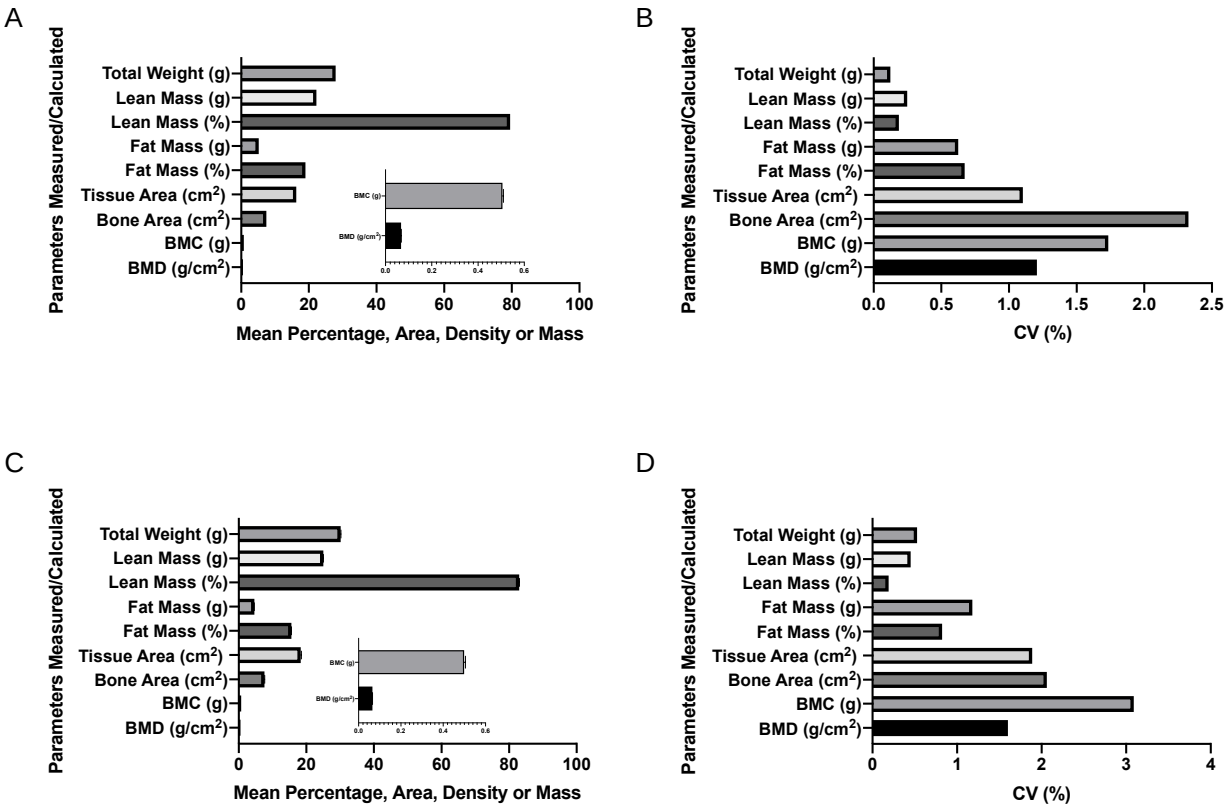


*Figure 1. Mean percentage, area, density or mass with calculated SEM and CV (%) for male mouse, measured 6 times. A and B) Non-repositioned, C and D) Repositioned.*

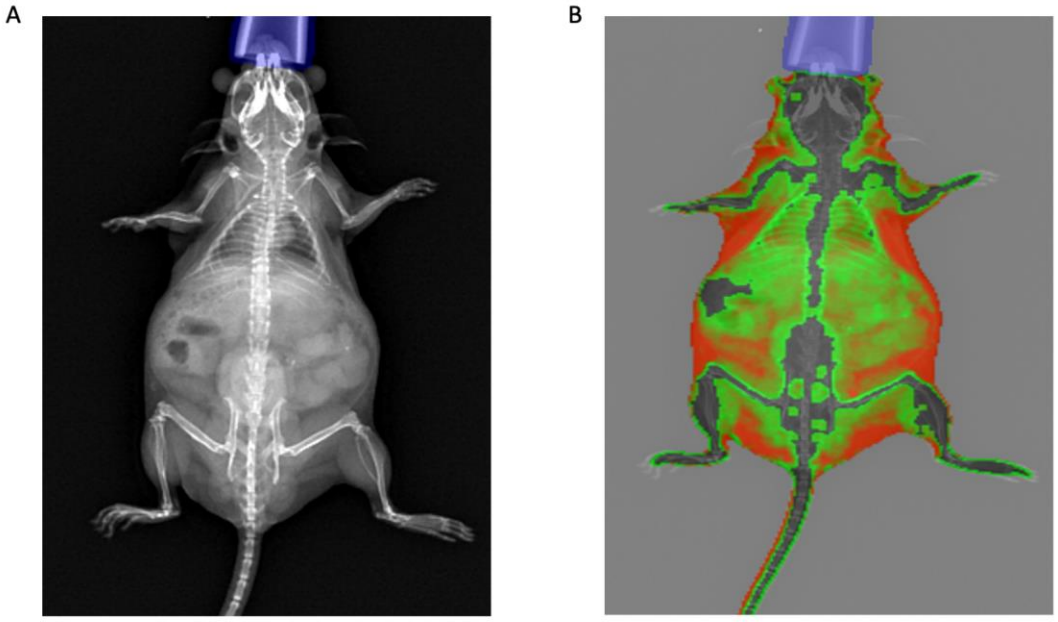


*Figure 2. Representative male mouse images. A) Standard x-ray attenuation image of male mouse. B) Lean (green) versus fat (red) mass image.*

The male rat was either non-repositioned or repositioned and the DXA measurements were acquired 7 times (Figure 3). CV values in all cases are equal or below 1.5% for the male rat. SEM values were calculated (Figure 3, A and C) and were minimal in all measurements.

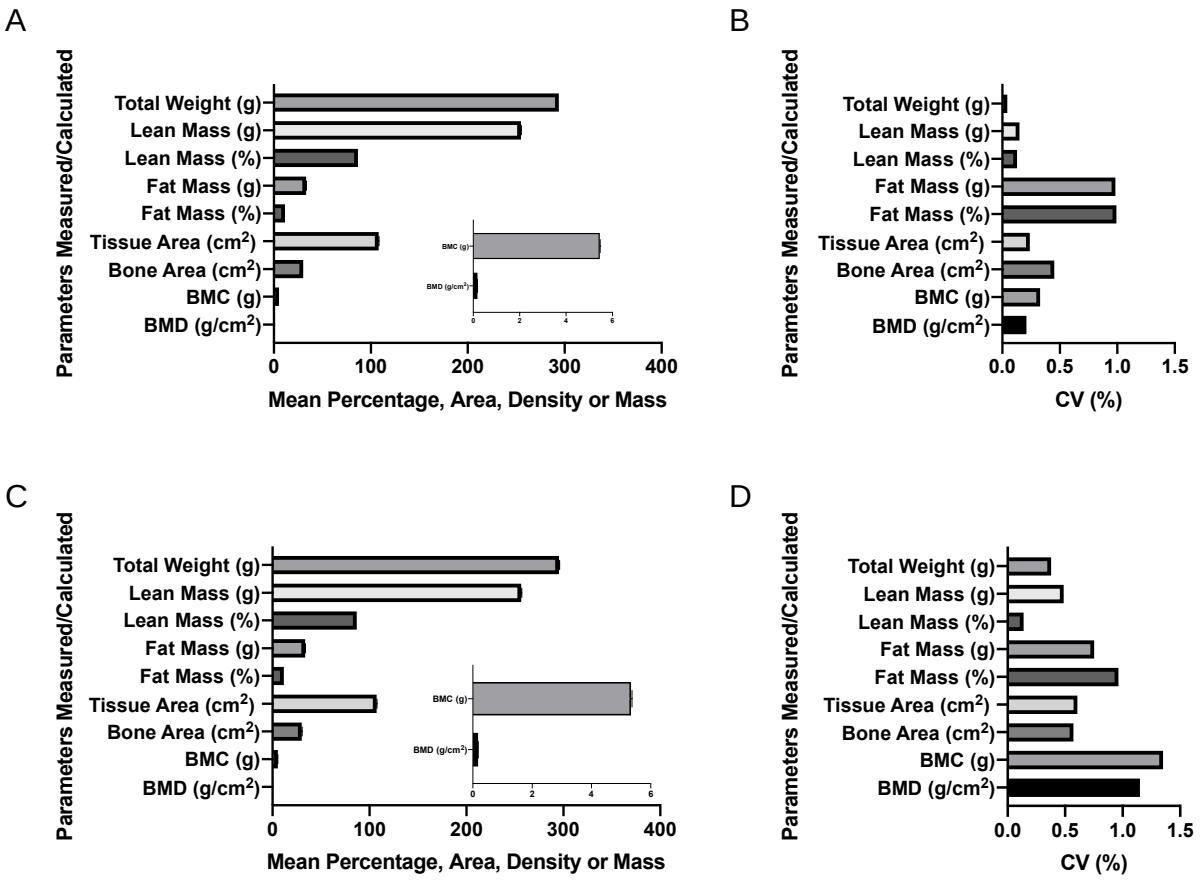


*Figure 3. Mean percentage, area, density or mass with calculated SEM and CV (%) for male rat (n = 7). A and B) Non-repositioned, C and D) Repositioned.*

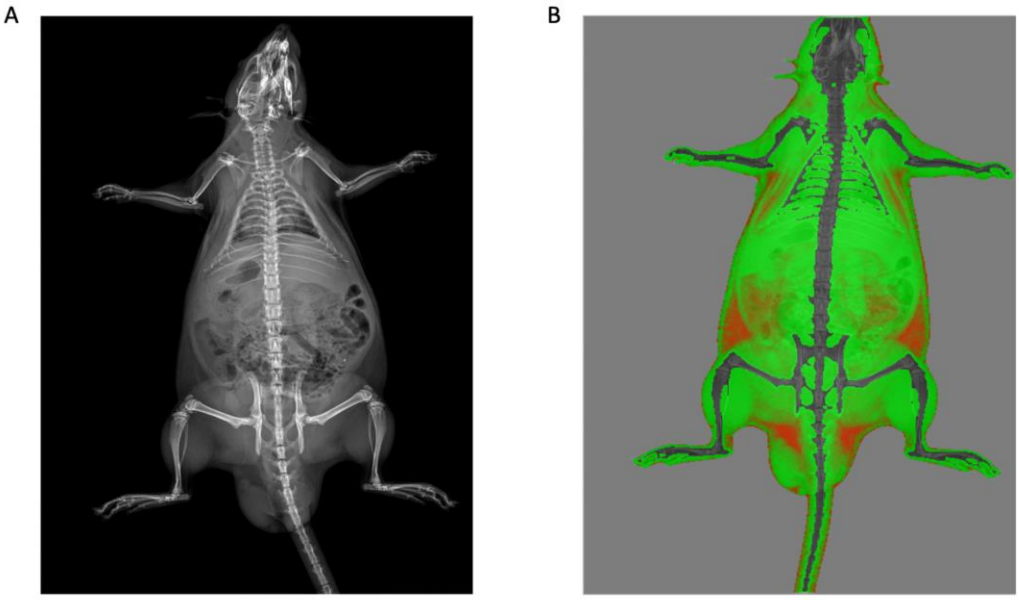


*Figure 4. Representative male rat images. A) Standard x-ray attenuation image of male mouse. B) Lean (green) versus fat (red) mass image.*

## Discussion and Conclusion

The data in this validation study shows a high level of reproducibility for the various measurements performed using the iNSiGHT DXA system. This reproducibility was achieved when the animal's position was maintained, as well as when the animal was re-positioned between measurements. This reproducibility is important when considering non-invasive longitudinal studies designed to look for small changes in body composition between animal groups, for which the iNSiGHT system was specifically designed. With low variability comes increased accuracy, and increased power in the resulting study data.

This data shows that the iNSiGHT system is well suited to measure the small changes expected in body composition over time in a variety of small animal models, and with the accuracy and reproducibility of the data the system is capable of reliably measuring small differences between control and study animals. Additionally, the iNSiGHT is well suited for longitudinal studies as the radiation dose is minimal, at 0.66mGy per scan, meaning the effects of multiple imaging timepoints would not affect the outcome of the study. Scans take 25 seconds to complete, allowing for high throughput of animals, with analysis available on the system or offline.