



## Variability of in vivo reference point indentation in skeletally mature inbred rats.

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[Allen, Matthew R.](#) ; [Newman, Christopher L.](#) ; [Smith, Eric](#) ; [Brown, Drew M.](#) ; [Organ, Jason M.](#)



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## Abstract:

Reference point indentation (RPI) has emerged as a novel tool to measure material-level biomechanical properties in vivo. Human studies have been able to differentiate fracture versus non-fracture patients while a dog study has shown the technique can differentiate drug treatment effects. The goal of this study was to extend this technology to the in vivo measurement of rats, one of the most common animal models used to study bone, with assessment of intra- and inter-animal variability. Seventy-two skeletally mature male Sprague-Dawley rats were subjected to in vivo RPI on the region between the tibial diaphysis and proximal metaphysis. RPI data were assessed using a custom MATLAB program to determine several outcome parameters, including first cycle indentation distance (ID-1st), indentation distance increase (IDI), total indentation distance (TID), first cycle unloading slope (US-1st), and first cycle energy dissipation (ED-1st). Intra-animal variability ranged from 13-21% with US-1st and Tot Ed 1st-L being the least variable properties and IDI the most highly variable. Inter-animal variability ranged from 16% (US-1st) to 25% (ED-1st 31 and IDI). Based on these data, group size estimates would need to range from 9-18/group to achieve sufficient power for detecting a 25% difference in a two-group experiment. Repeat tests on the contralateral limb of a small cohort of animals (n=17) showed non-significant differences over 28 days ranging from -6% to -18%. These results provide important data on RPI variability (intra- and inter-animal) in rats that can be used to properly power future experiments using this technique.

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