THE OSTEONECIC POTENTIAL OF REST-INSERTED LOADING

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INTRODUCTION

The use of mechanical stimuli to maintain or augment bone mass has shown considerable promise in animal models of bone loss [1], but its potential application in human subjects has met with limited therapeutic success [2]. This is, in part, because the high-magnitude, high-impact forms of exercise typically associated with bone accretion defy implementation in the elderly due to difficulty with compliance [3], and in the young due to deleterious effects upon longitudinal bone growth [4]. One possible solution may lie in our recent observation that the insertion of a rest period between loading cycles enhances osteoblastic activation elicited by low-magnitude mechanical loading by over 6-fold [5]. However, application of this concept in human subjects will first require demonstration of enhanced bone formation in a model wherein the mechanical stimulus is superposed upon normal functional activity. In this study, we have therefore utilized our newly developed non-invasive murine tibia loading device [6] to examine the hypothesis that rest between load cycles enhances the osteogenic potential of low-magnitude mechanical loading.

METHODS

Three groups of adolescent female C57BL/6j mice (age: 10 wk) underwent exogenous mechanical loading of their right tibia utilizing the non-invasive murine tibia loading device (Fig 1). The right tibiae of the first group of subjects were subject to a identical 1Hz load waveform as the low-magnitude group with the following exceptions: the right tibiae were loaded for only 10 c/d and a unloaded rest period of 10-s was inserted between each of the 10 load cycles (rest-inserted). All animals were then allowed 3 additional weeks of normal cage activity prior to sacrifice. Thus, the overall daily loading duration for animals in all groups were identical (100 s). A subset of animals in each group received calcine (ip: 15 mg/Kg) on days 5 and 21 to detect activation of the periosteal surface, a site where bone formation is structurally most beneficial. Upon sacrifice, the right (experimental) and left (intact contralateral control) tibiae were disarticulated and dissected of soft tissue. Mid-diaphyseal cross sections were then prepared (100 µm) and mounted unstained for evaluation of cross-sectional areal properties and dynamic indices of bone formation. Digital images of sections were obtained using a epi-fluorescent microscope and primary measures of cortical bone area (Ct.Ar), periosteal bone surface (BS), single labeled surface (s.LS), double labeled surface (d.LS), and interlabel width (Ih.L.Th) were obtained. Cortical bone area was expressed as the ratio of right to left areas (rCt.Ar), and mineralizing surface (MS), mineral apposition rate (MAR) and bone formation rate (BFR) were calculated per standard procedures [7]. Non-parametric statistics were utilized to detect significant differences between measures.

RESULTS

One week of loading followed by three weeks of cage activity increased rCt.Ar within each group (low-magnitude: 6.5 ± 5.5%, high-magnitude: 4.6 ± 2.6%, rest-inserted: 16.9 ± 5.9%) but significant differences were not detected between groups (high vs low: p = 0.42; rest vs low: p = 0.08). Low-magnitude loading did not increase BFR vs control bones (0.04 ± 0.04 vs 0.02 ± 0.02 µm2/m2/d, p = 0.22). In contrast, the high-magnitude (0.3 ± 0.1 µm2/m2/d, p < 0.01) and rest-inserted loading (0.4 ± 0.2 µm2/m2/d, p = 0.01) significantly increased BFR (Fig 2). Likewise, MAR elicited by both high-magnitude (1.1 ± 0.2 µm/d, p < 0.01) and rest-inserted loading (1.4 ± 0.6 µm/d, p < 0.01) were significantly increased. Last, only rest-inserted loading significantly altered MS vs controls (rest-inserted: 28.3 ± 1.5%, p = 0.05; high-magnitude: 25.3 ± 3.4%, p = 0.06; low-magnitude: 18.1 ± 4.6%, p = 0.5; control: 18.2 ± 3.0%).

DISCUSSION

In growing adolescent animals, the ability of loading protocols to augment existing bone formation beyond the background level of activity in control bones can be used to evaluate the osteogenic potential of each loading regimen. Within this context, the substantial bone formation induced by high-magnitude loading and the inability of low-magnitude loading to alter bone formation serve as positive and negative controls for the loading device itself. In support of our hypothesis, insertion of a 10-s rest period between each of 10 load cycles induced significant bone formation in the murine tibia mid-diaphysis. This contrasts with the lack of response elicited with 10 times more cycles of loading at the same strain magnitude (and strain rate) in the low-magnitude group. Surprisingly, the osteogenic potential of 10 cycles of rest-inserted loading was equivalent to that induced using a standard 1 Hz waveform possessing twice the strain magnitude, twice the strain rate, and 10 times the cycle number. The strong osteogenic potential of short-term rest-inserted loading holds promise for optimizing long term protocols capable of significantly enhancing bone mass and morphology. If so, low-magnitude rest-inserted loading would present a readily implemented (e.g., via modified tai chi), viable alternative to high-impact, high-magnitude loading as a therapeutic strategy to combat skeletal fragility.


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