Introduction: We have previously reported an age-related loss of joint innervation in a mouse model of spontaneous osteoarthritis (1). This loss seemed to be more significant in the population of large caliber, presumably mechanoreceptor neurons. A loss of large caliber joint afferents could result in degenerative changes in articular cartilage due to the failure of reflexes appropriate and necessary to protect articular surfaces from abnormal loads by the appropriate timing of contractions of the muscles acting around the joint (2, 3). Many neuronal networks have an inherent redundancy or compensatory capacity, such that functional deficits are not evident until losses exceed a certain threshold (e.g. the nigrostriatal system in Parkinson’s disease). Accordingly, we hypothesized that there would be a higher likelihood of degenerative change in joints with a loss of innervation only below a certain threshold.

Purpose: To correlate the histologic appearance of the knee joint with the number of identifiable sensory neurons supplying the joint in cohorts of mice at three different ages chosen to span the mean lifespan of the mouse.

Methods: All experiments were carried out in accordance with the guidelines of the Canadian Council on Animal Care. C57BL/6 isogenic mice were obtained from the colony held by the U.S. National Institute for Aging at 26, 52 and 96 weeks of age (n=6 in each group). Each mouse was injected with 2% Fluoro-Gold (FG) into one or both knees. After 2-5 days the lumbar dorsal root ganglia (DRGs) were harvested and serially sectioned for determination of the total number of labelled joint afferents according to our published protocol (1). The corresponding knee joints were decalcified, embedded in paraffin and the complete set of serial sagittal sections collected. Alternate sections were stained with H & E or haematoxylin, Fast green and saffronine-O for histological determination of the number and grade of degenerative lesions according to the the published criteria of Maier and Wilhelmi (4). Lesions were weighted from 1 to 4 by these criteria and each joint assigned a numerical score by examining the complete set of serial sections through the tibio-femoral articulation of each joint and summing the joint assigned a numerical score by examining the complete set of serial sections through the tibio-femoral articulation of each joint and summing the total number of lesions, weighted by grade of severity. We then plotted the OA score of each knee as a function of the number of DRGs for that knee (Fig 1).

Results: Each point in figure 1 below indicates the data for one knee joint from one mouse. The dotted line represents an arbitrary threshold of one hundred joint afferents, representing 40% of the average number of joint afferents supplying the knee joint at 8 weeks of age in this model. Below this arbitrary threshold, the prevalence of advanced degenerative change seems to increase.

Discussion: The results reconfirm that in the C57BL6 mouse, the numbers of knee joint afferents decrease significantly with aging, consistent with our previous work showing that there was an average 60% loss of the total number of knee joint afferents over the life span of the mouse (1). In this experiment, significant degenerative change was not seen in any knee joints with an afferent nerve supply greater than 100 neurons. Conversely, in 52 week mice, knee joints with fewer than 100 joint afferents did not typically show advanced degenerative change. If a loss of joint innervation is a causal factor in OA then this loss would be expected to precede the development of advanced degenerative change. This supports the hypothesis that this age-related loss of joint innervation (below the threshold of 40% of the initial complement) may be a permissive or promoting factor in the spontaneous development and progression of osteoarthritis in this strain of mouse but does not exclude other possibly important factors.

One of the 96 week old mice did not show much degenerative change in either knee (asterisks in Figure 1). Interestingly, one of the knee joints of this mouse had a well-preserved innervation, the other did not. Yet neither joint developed advanced degenerative change. Prior to the design of this study, we did not consider the possibility that a better-innervated knee joint might be able to exert a protective influence on the contralateral knee of the same animal. For this reason we did not examine both knees of all animals. No other animal that was examined bilaterally showed a similar disparity of joint innervation. The C57BL6/NNia mouse is an inbred strain in which individuals are all genetically virtually identical. Reasons for the observed differences in losses of joint afferents and severity of articular cartilage degenerative changes between individuals are not readily apparent, but are not likely to be due to genetic differences.

References