INTRODUCTION: Excessive spine loading due to tennis, swimming, golf or contact sports can exert significant stress on the intervertebral disc (IVD), potentially leading to degeneration and pain. We simulated a common sport movement consisting of rotations about all three anatomical axes (i.e. extension, lateral bending and torsion). The simulation was performed in a novel bioreactor that enables testing of \textit{ex vivo} whole organ IVDs under multiaxial loading in a biologically controlled environment [1]. In this study, we assessed the impact of different magnitudes of this complex motion [2] on IVD physiology and the specific contribution of bending motions alone to the observed effects.

METHODS: Based on the previously described procedure [1], bovine caudal whole IVDs were isolated from 10 donors and subjected to 3 different loading protocols: (Group 1), Extension 0-3°, Lateral Bending 0-3° and Torsion 0-2°, (Group 2) Extension 0-6°, Lateral Bending 0-6° and Torsion 0-4°, (Group 3) Extension 0-6° and Lateral Bending 0-6°. All protocols were run daily for 2h at 0.3 Hz for 14 days. During the loading and free swelling recovery, IVDs were kept in the culture medium in a sterile biologically controlled environment (Fig.2). Based on the previously described procedure we showed that torsion has a stronger GAG and NO release effect than rotations at lower magnitudes (Fig.2A,E). Furthermore, it was accompanied by reduced collagen type-II immunostaining at the interface between AF and NP (Fig.2D) and disintegration of NP structure (Fig.2A). Phenotype analyses indicated that all groups, irrespective of magnitude and omitting torsion, have upregulated collagen genes MMP1 and MMP13 in AF and NP, with particularly high values in group 1 (38- and 316-fold increase for MMP1 and 69- and 238-fold increase for MMP13, respectively). Based on a macroscopical evaluation of transverse sections from the IVD center, all three loading regimes caused high cell death in NP and iAF (Fig.2E) and cell clustering in NP typical of IVD degeneration (Fig.2B,C), aligning with the changes of extracellular matrix (ECM) observed in these regions.

DISCUSSION: We showed that a combination of two or more cyclic rotations about the anatomical axes induces high cell death and catabolic response at the IVD center (i.e. NP) and the interface with AF, possibly caused by detrimental mechanical stresses at these locations. This effect occurred even when the loading was run at a physiological frequency of 0.3 Hz and despite a minimally reduced IVD height. However, data from diurnal analyses suggest that torsion at 4° additionally to backward and lateral bending at 6° induces a stronger GAG and NO release than rotations at lower magnitudes, making the IVD prone to mechanical impairment caused by inflammatory response and ECM degradation and release.

SIGNIFICANCE/CLINICAL RELEVANCE: This pioneering study in a new generation of bioreactors brought a new perspective on the potentially detrimental effect of intensive sport motion on IVD homeostasis, making the spine prone to injury, impairment and/or low back pain. It can further help optimize protocols for spine health maintenance during exercise based on the type of movement and intensity of daily loading.


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IMAGES AND TABLES:

Fig.1. Accumulative GAG (A) and NO (B) release in the IVD cell culture medium from 3 different loading regimes.

Fig.2. Representative histology sections from group 2: reduction or loss of red positive GAG staining in NP (A) and iAF (A,E), cell clusters (arrow) in NP (B,C), reduced collagen type II immunohistostaining in iAF (D), viable cells stained in blue in outer AF and innermost NP only (F).