Cellular Behavior In A Partial-Thickness Cartilage Defect Under Hydrostatic Pressure and/or Strain Mimicking Weight-Bearing and Joint-Loading

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Disclosures

The authors have no disclosures.

INTRODUCTION

- Partial-thickness cartilage defects can lead to further chondral degeneration and osteoarthritis;
- At the cellular level, the influence of mechanical loading on this chondral degeneration is still to be determined;
- Chondrocyte metabolism, differentiation and proliferation are affected by different mechanical conditions;
- By mimicking knee bending and normal gait pressures, **dynamic bending strain (BS) and cyclic hydrostatic pressure (HP)** can reliably replicate the physiological mechanical loading that acts in the knee during normal joint movements.

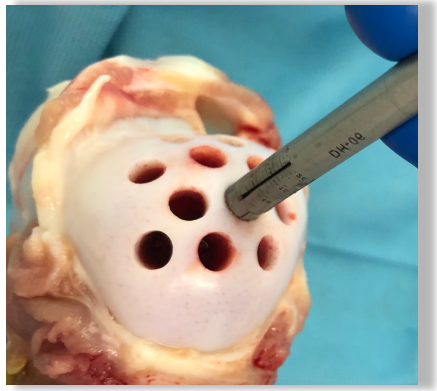
Aim:

In this study, we aimed to characterize the cellular behavior in a partial-thickness cartilage defect model under cyclic hydrostatic pressure (HP) and/or arc-bending strain (BS)

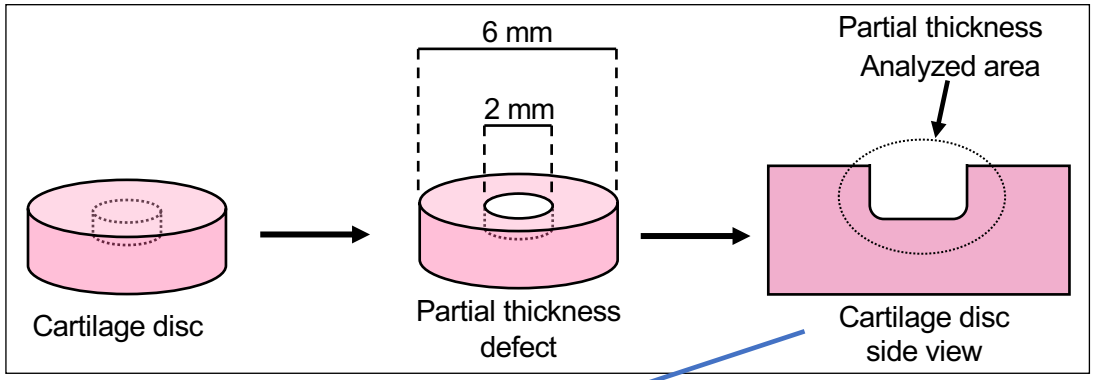
We **hypothesize** that chondrocytes surrounding the cartilage defect proliferate under compressive stress/joint loading in the knee cartilage

METHODS

Bovine cartilage were used in this study. Round articular cartilage discs (6-mm) were harvested from bovine humeral head

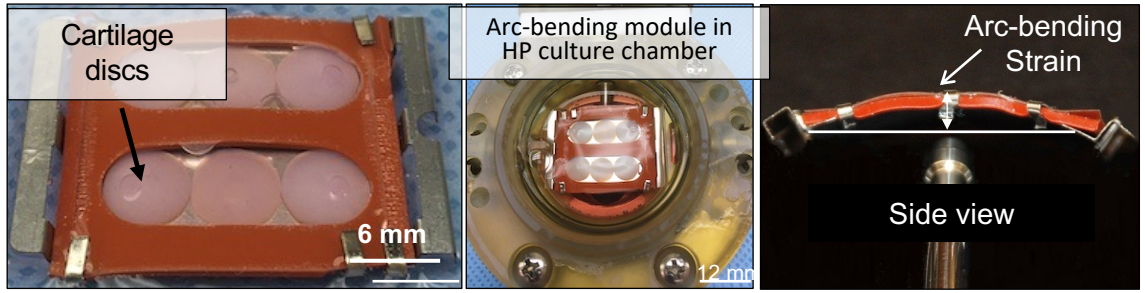


A partial-thickness defect (2 mm in diameter x 1 mm in depth) was made at the center of each disc



Discs were randomly allocated to 4 culture conditions

Group	Culture Conditions
Atmospheric pressure (AP)	Static conditions at 37°C atmospheric pressure (no HP)
Arc-bending strain (BS)	Continuous BS alone with a frequency of 0.5 Hz, at 37°C
Hydrostatic Pressure (HP)	HP at 0-0.5 MPa, 0.5 Hz, 37°C
HP/BS	Combined HP/BS, frequency of 0.5 Hz, at 37°C



Arch-bending module **(BS)** and pressure culture system applying cyclic hydrostatic pressure **(HP)** were used to replicate knee loading and weight bearing, respectively

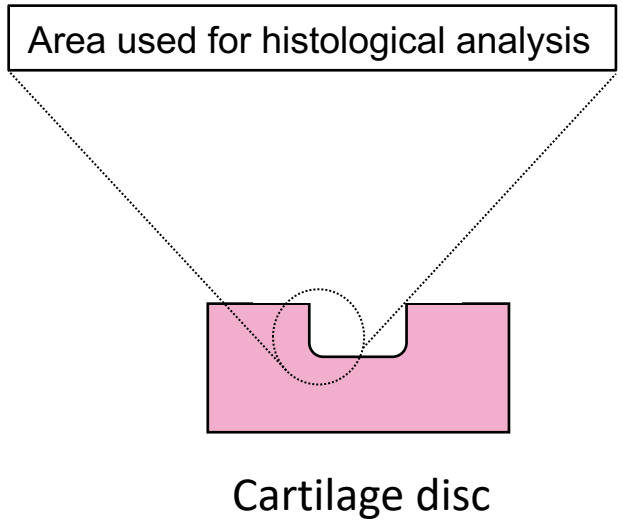
METHODS

Discs were harvested (n=8/group) at days 6 and 9 for histological evaluation

Evaluation:

- Cellular viability - Live/Dead staining
- Cartilage matrix and anucleate cells - Safranin-O (Saf-O)/ hematoxyline
- Cell proliferation - Proliferating cell nuclear antigen (PCNA) antibody
- Tissue degeneration - Matrix metalloproteinase (MMP)-13 antibody

Percentage of live/dead cells, anucleate cells, PCNA-positive and MMP-13 positive cells were counted



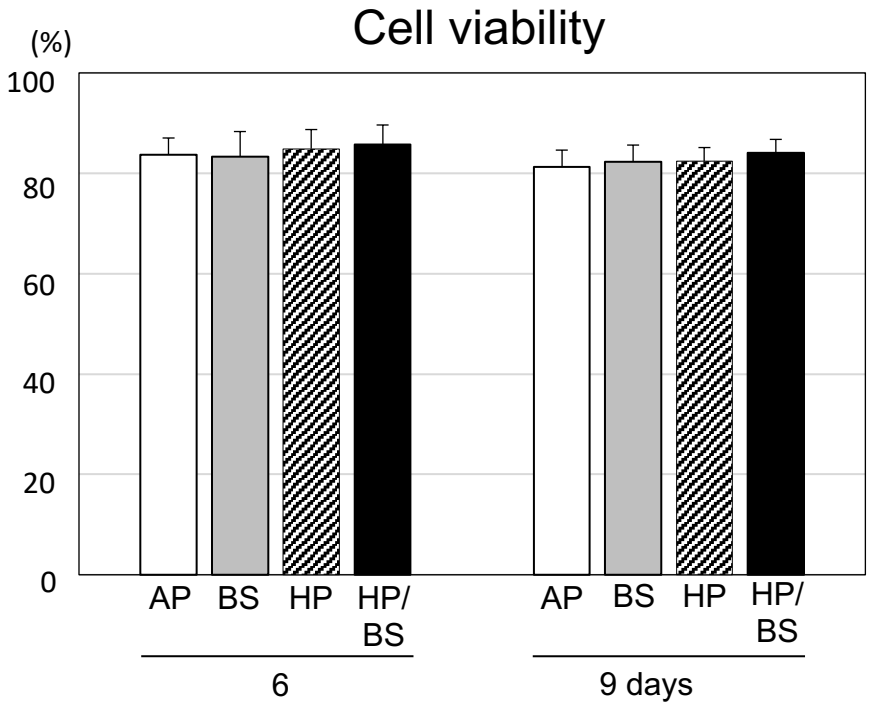
Statistical analysis

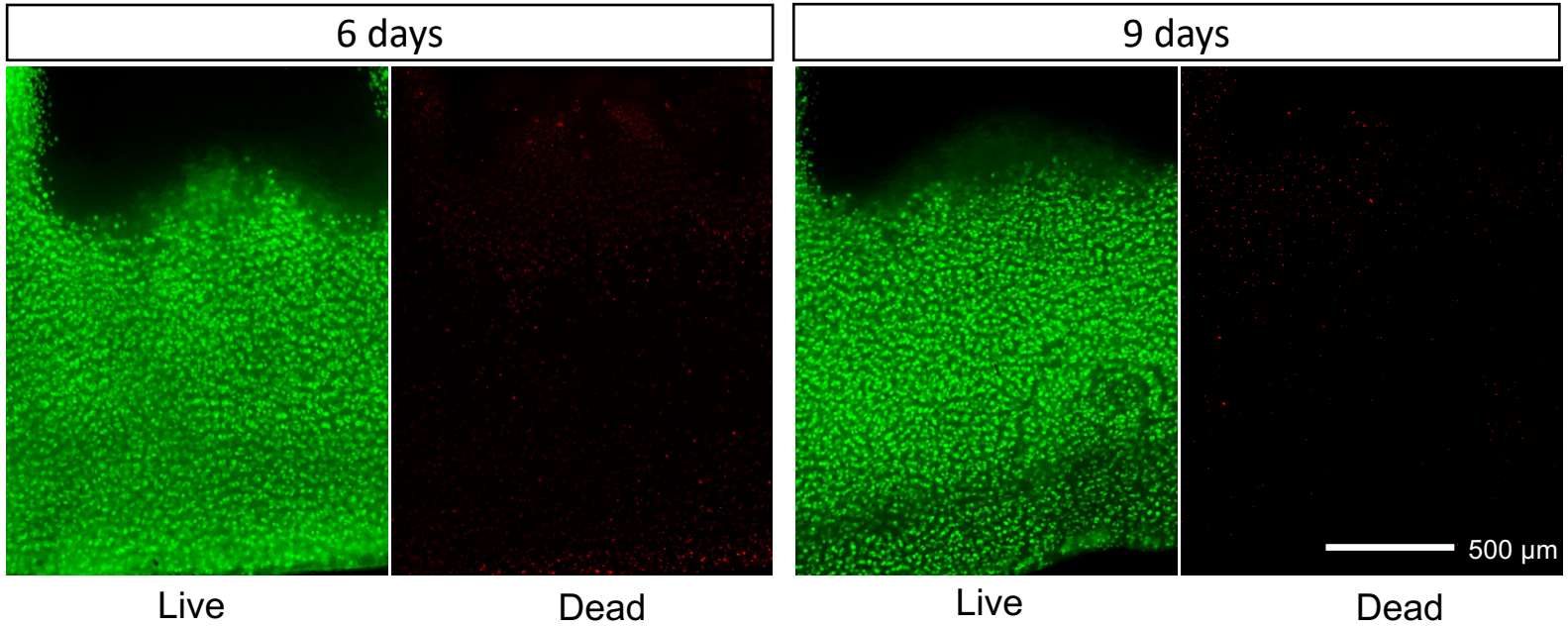
Two-way analysis of variance followed by Tukey's post hoc tests were used for statistical analysis.

Statistical significance was set at $p < 0.05$.

RESULTS

Cell viability remained high in all groups (>80%) over 9 days of culture

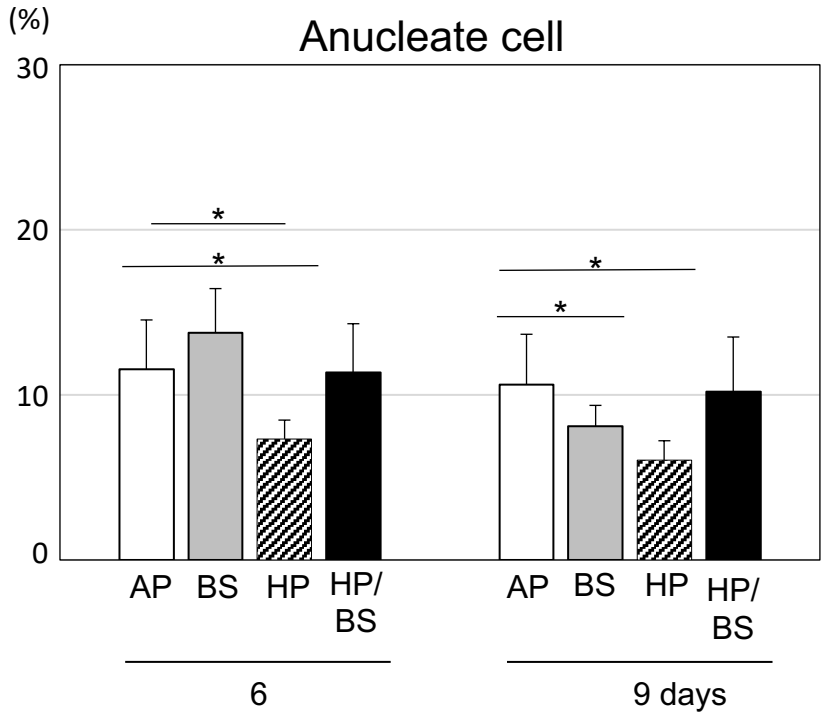


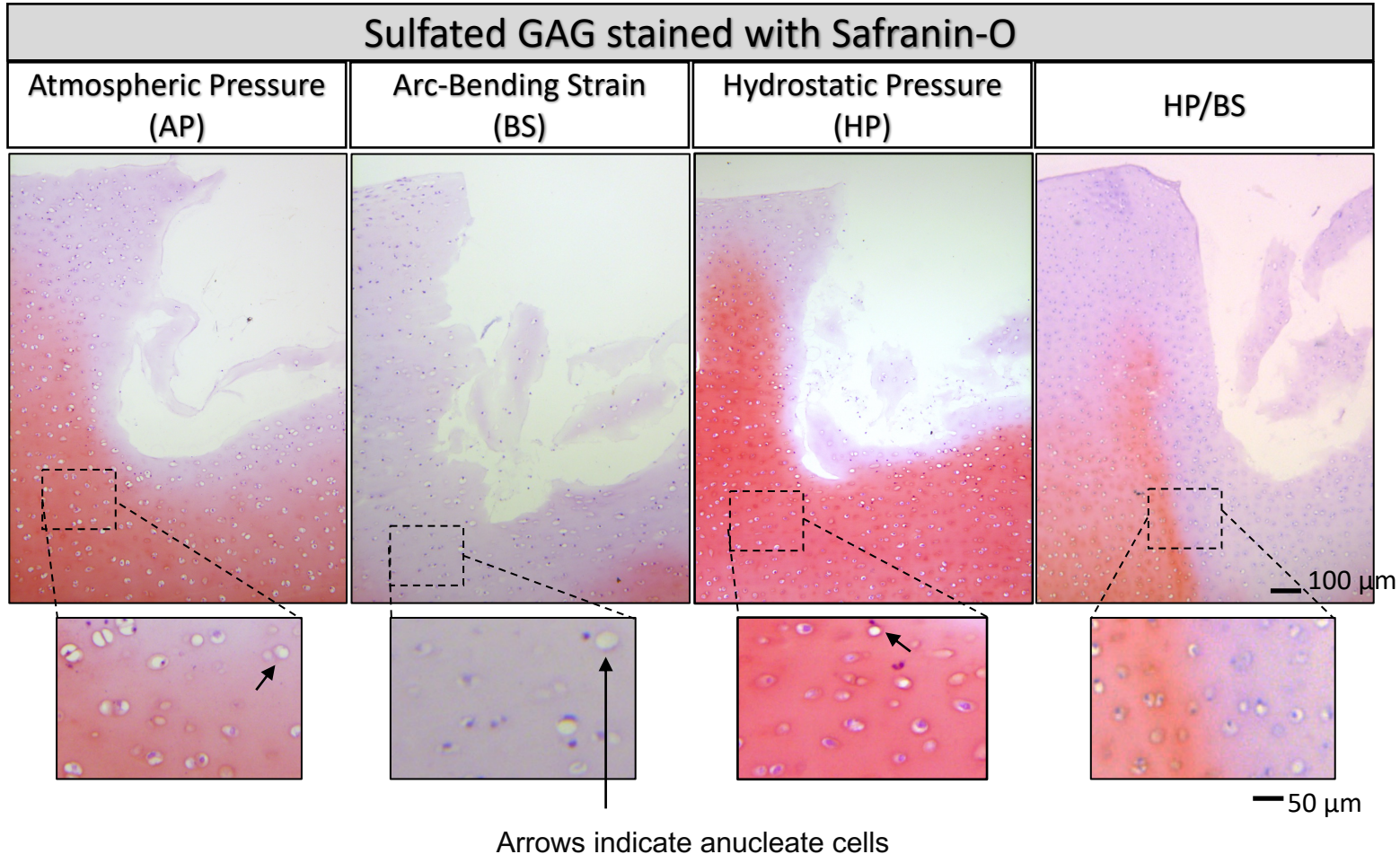


Green=Live cells / Red= dead cells

RESULTS

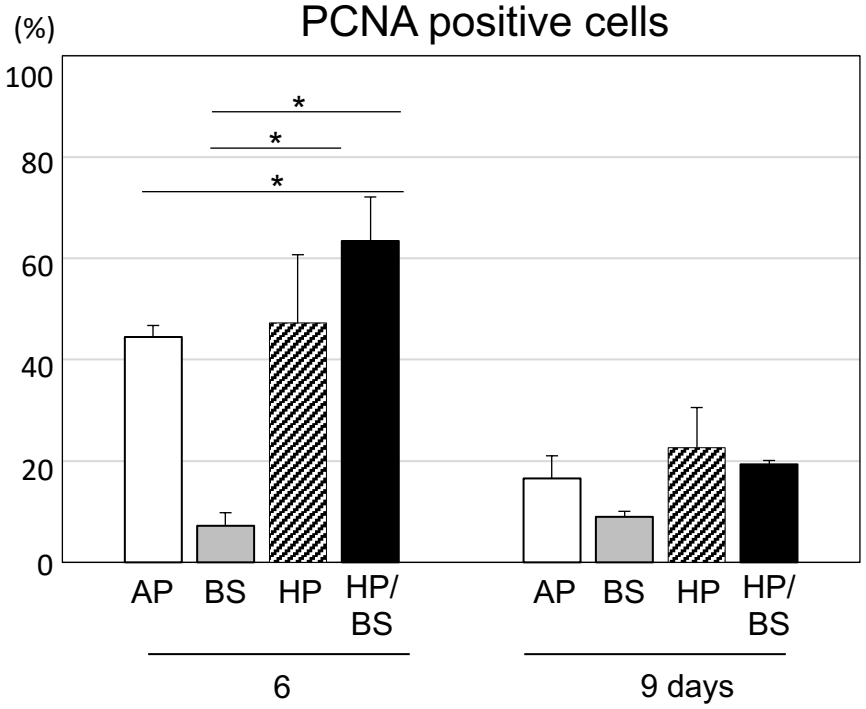
HP showed less anucleate cells in comparison to the other conditions

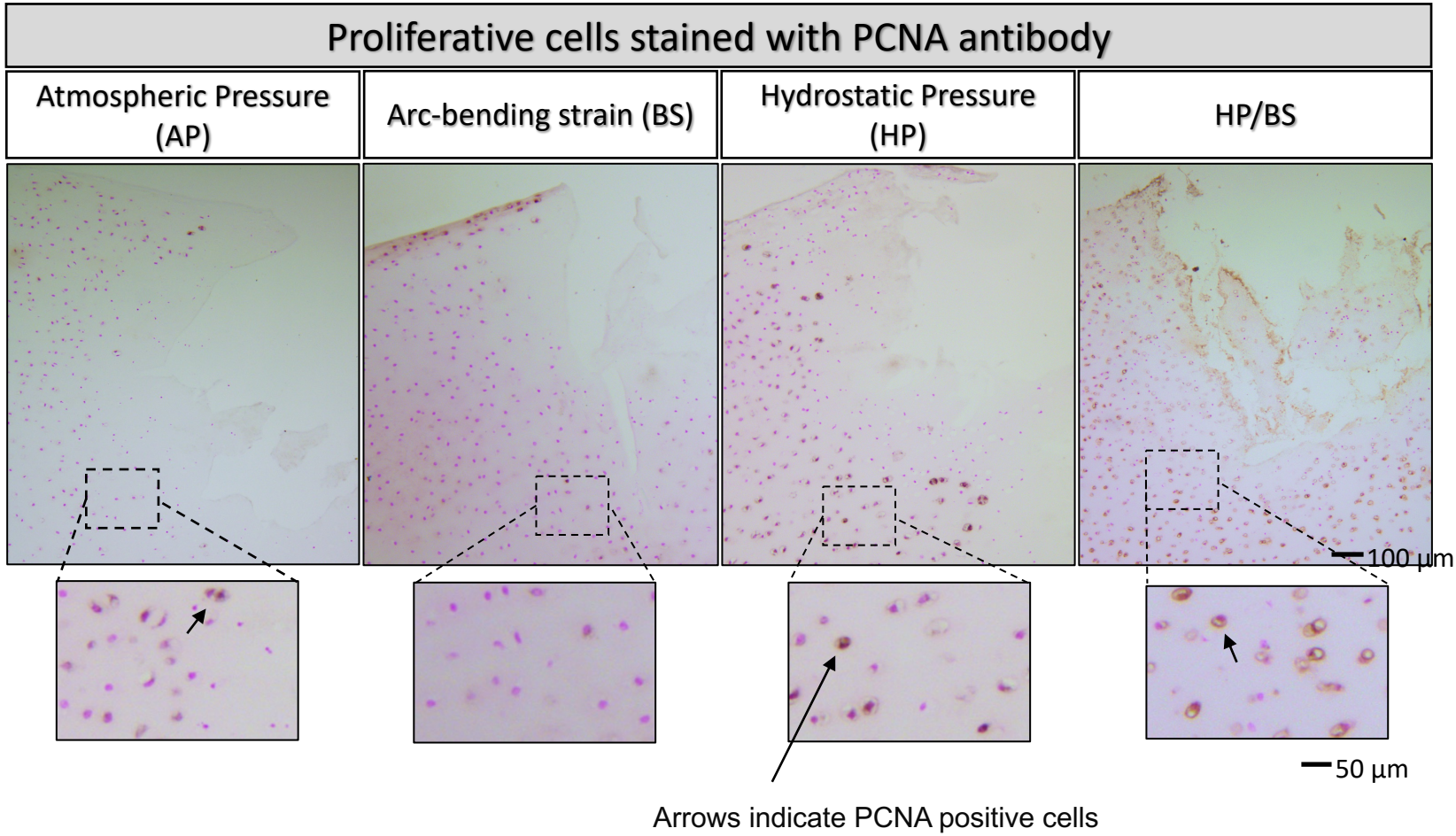




RESULTS

HP and HP/BS led to higher PCNA positive cells after 6 days of exposure

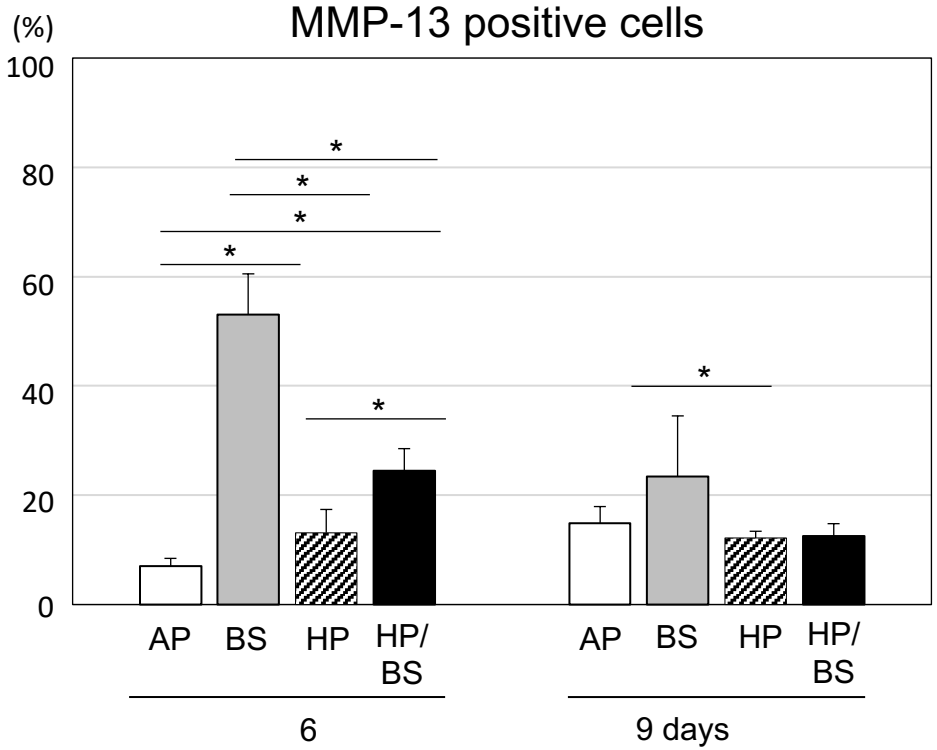


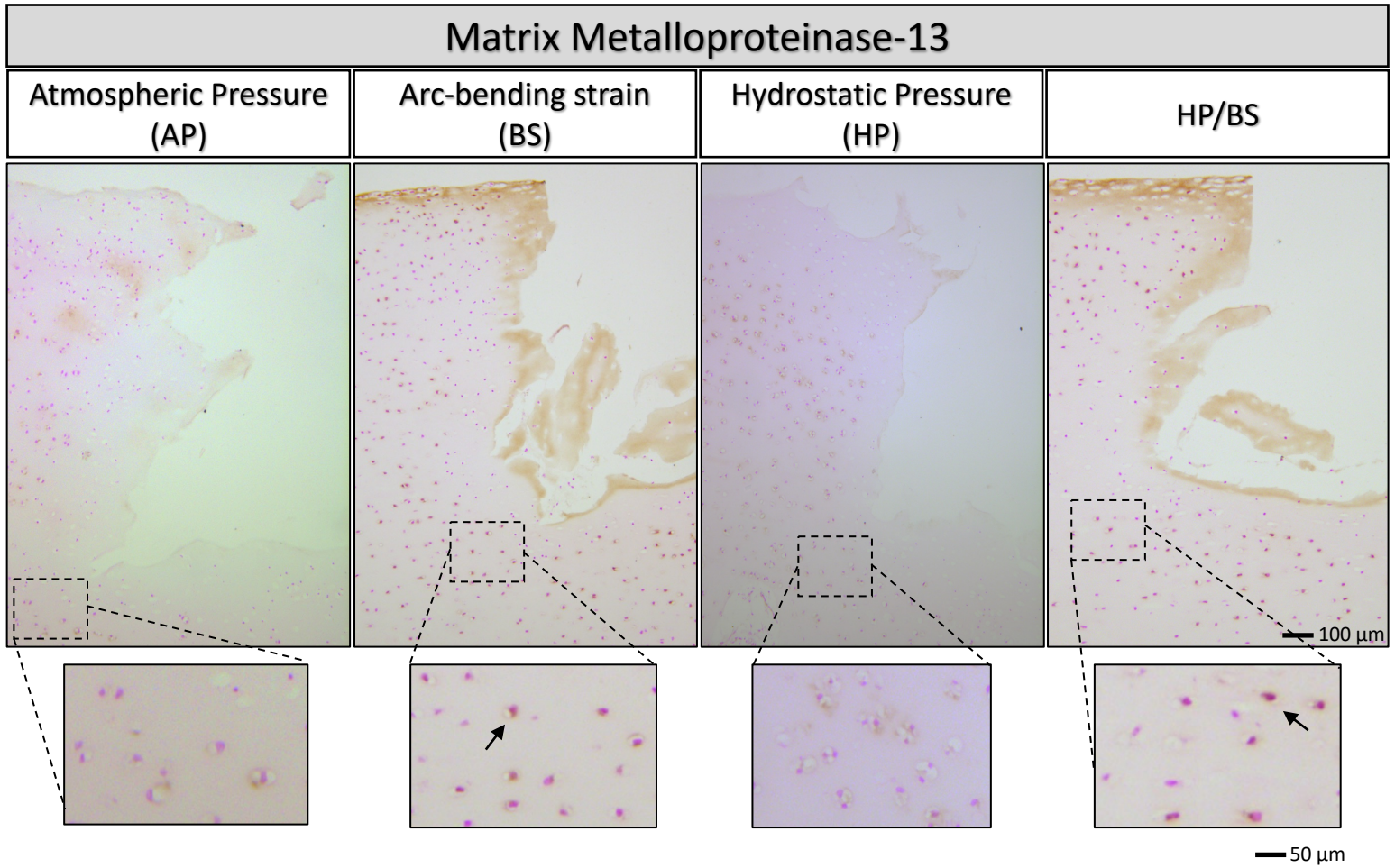


Arrows indicate PCNA positive cells

RESULTS

MMP-13 positive cells was higher under BS





Arrows indicate MMP-13 positive cells

DISCUSSION

The percentage of PCNA and MMP-13 positive cells among groups showed opposite trends:

- **Bending Strain (BS) alone more strongly suppressed cell proliferation** and stimulated enzymatic degeneration (**MMP-13**).

- However, **combining Bending Strain with Hydrostatic Pressure (HP/BS) stimulates cell proliferation and suppressed MMP-13**.

    - Since HP/BS prevented suppression of cell proliferation and inhibition of catabolic enzyme activation, compressive loading (HP/BS) in articular cartilage seems to be a key factor for healthy cartilage homeostasis.

DISCUSSION

Clinically, **HP** has the capability of mimicking **weight bearing** while **BS alone** may mimic **non-weight bearing joint movement.**

- The association **HP/BS** positively affected the chondrocyte activity at the articular cartilage defect, increasing cell proliferation and reducing catabolic response, which was not observed with BS alone.

- This effects was significantly higher after 6 days of **HP/BS** but not after 9 days of **HP/BS**.

- These findings support a hypothesis that knee range of motion alone during the early rehabilitation post-cartilage repair treatments may not be enough to properly stimulate an adequate healing response.
  - The concomitant use of weight-bearing would be required to improve the cartilage healing capacity propelled by the post-operative rehabilitation.

CONCLUSION

While **BS alone** suppressed cell proliferation and stimulated ECM degradation, **combined HP/BS** overcame these responses, suggesting the key role of compressive loading (HP/BS) in maintaining a healthy articular cartilage.

We established a dynamic cartilage defect model with bending strain and cyclic hydrostatic pressure without unrecoverable tissue deformation. This model can be used for evaluating regenerative or degenerative changes in articular cartilage surface under weight-bearing and joint-loading. Since little information is known about early changes after surface injuries, this model holds potential in the development of repair strategies and postoperative rehabilitation protocols.

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