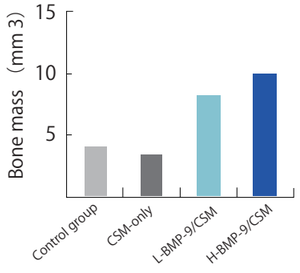
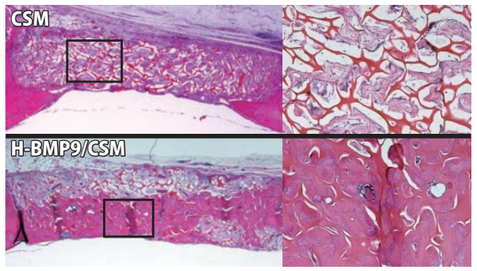
Collagen for Cell Culture
AteloCell®

Example 5 Sustained release of rhBMP-9 and osteogenesis using MIGHTY

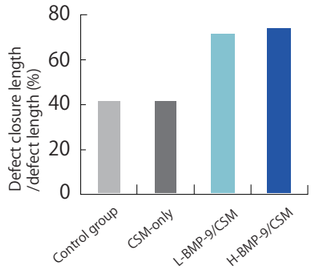
(Nakamura T, Noguchi K, Kagoshima University Graduate School of Medical and Dental Sciences, Department of Periodontology)



Bone mass evaluation 8 weeks after transplantation



H&E staining image 8 weeks after transplantation
(low-power field image and enlarged image within the black frame)



Histometric evaluation 8 weeks after transplantation

1 μg of rhBMP-9 (L-BMP-9) or 5 μg of rhBMP-9 (H-BMP-9) was added to MIGHTY (CSM) and implanted it into the rat skull defect. 8 weeks after transplantation, a significant increase in bone mass was observed in both rhBMP-9/CSM groups compared to the control group and the CSM alone transplantation group. In histological evaluation, connective tissue dominated in the CSM-alone transplant group, whereas new bone formation was observed within the CSM in both rhBMP-9/CSM groups. Additionally, both rhBMP-9/CSM groups showed a significant difference in defect closure rate compared to the control group. (Reference 1)

Reference

1. Nakamura T, *et al.* Osteogenic potential of recombinant human bone morphogenetic protein-9/absorbable collagen sponge (rhBMP-9/ACS) in rat critical size calvarial defects. (2017) *Clin Oral Investig.* 21(5):1659-1665.
2. Kajiyama S, *et al.* Bone formation by human umbilical cord perivascular cells. (2015) *J Biomed Mater Res A.* 103(8):2807-2014.
3. Nishio A, *et al.* Chondrocyte differentiation of human buccal fat pad-derived dedifferentiated fat cells and adipose stem cells using an atelocollagen sponge. (2015) *J Osaka Dent Univ.* 49(2), 185-196.
4. Haniu M, *et al.* A study on optimum orthodontic force using human periodontal ligament cells on 3D culture system. (2014) *Hokkaido J. Dent. Sci.*, 34：97-105.
5. Hara M, *et al.* Effect of strain on human dermal fibroblasts in a three-dimensional collagen sponge. (2014) *Cytotechnology.* Oct;66 (5):723-728.
6. Hatano Y, *et al.* Tumor associated osteoclast-like giant cells promote tumor growth and lymphangiogenesis by secreting vascular endothelial growth factor-C. (2014) *Biochem Biophys Res Commun.* 446(1):149-154.
7. Kanazawa T, *et al.* Biological responses of three-dimensional cultured fibroblasts by sustained compressive loading include apoptosis and survival activity. (2014) *PLoS One*. 9(8):e104676.
8. Shimomura K, *et al.* Cyclic compressive loading on 3D tissue of human synovial fibroblasts upregulates prostaglandin E2 via COX-2 production without IL-1β and TNF-α. (2014) *Bone Joint Res.* 3(9):280-288.
9. Ota K, *et al.* Optimal cyclic compressive loading promotes differentiation of 3D-cultured pre-osteoblasts. (2013) *J Osaka Dent Univ.* 47 (1):117-125.
10. Shiozaki Y, *et al.* Enhanced in vivo osteogenesis by nanocarrier-fused bone morphogenetic protein-4. (2013) *Int J Nanomedicine.* 8:1349-1360.

Cat. No.	Description	Quantity	Storage
KOU-CSM-25	Atelocollagen sponge Mighty	25 pcs/btl	room temprature
KOU-CSM-50	Atelocollagen sponge Mighty	50 pcs/btl	room temprature

World distributor

COSMO BIO USA.

2792 Loker Avenue West, Suit 101, Carlsbad, CA 92010 USA
Phone : +1 760-431-4600 / Fax : +1 760-431-4604
e-mail : support@cosmobiousa.com
web : https://www.cosmobiousa.com/

Please don't hesitate to contact us with any questions related to product selection or use.

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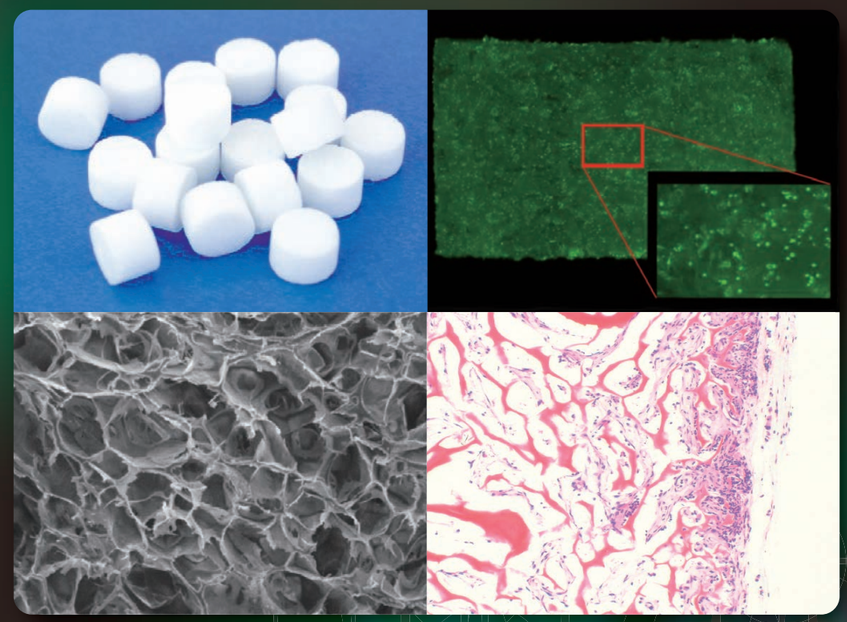
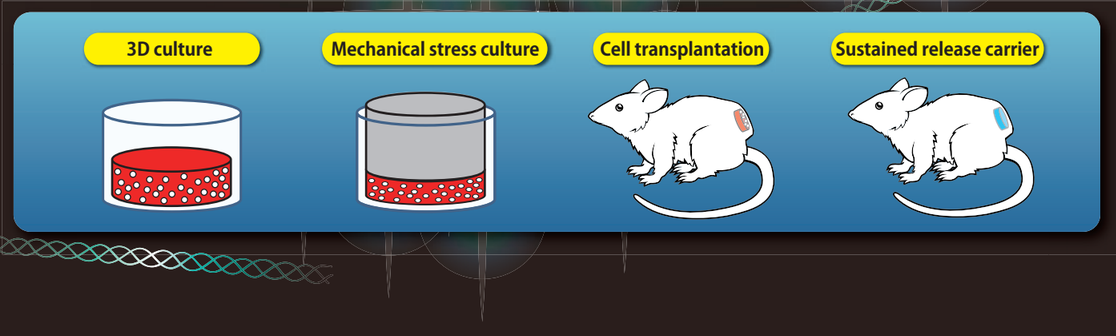
AteloCell® Collagen for Cell Culture

Atelocollagen Sponge, MIGHTY





Collagen for Cell Culture AteloCell®

Atelocollagen Sponge, MIGHTY CSM-25/CSM-50

Product summary

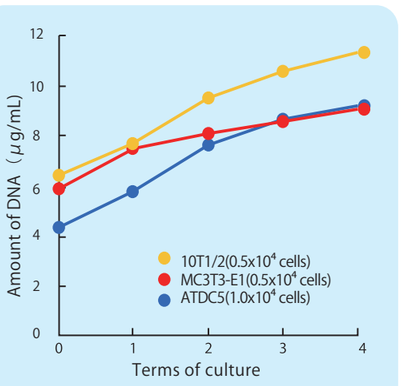
Atelcollagen Sponge MIGHTY is a collagen sponge that is primarily composed of atelocollagen and is strong enough to retain structural rigidity during repeated compressive loadings of 40 kPa. By culturing the seeded cells with mechanical stimulation (repetitive loading), cell function can be evaluated under conditions close to living organisms. It can also be used as a scaffold for cells in 3D culture and cell transplantation.

Applications

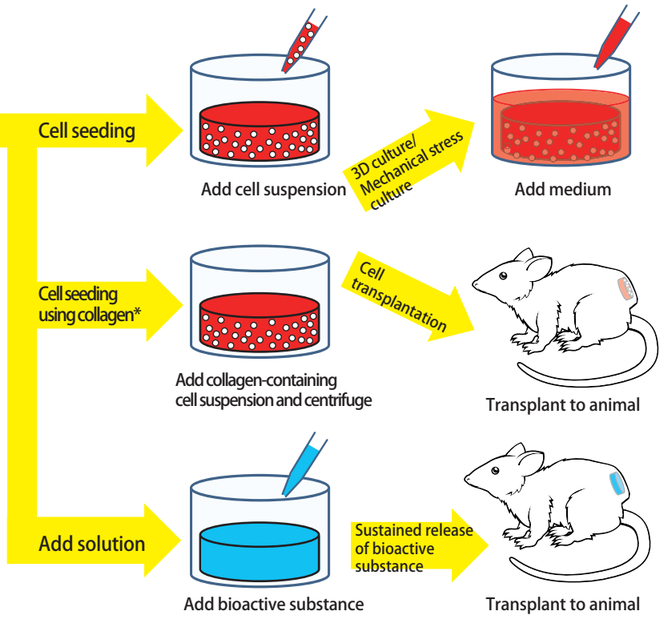
- 3D culture
- Mechanical stress culture
- Cell transplantation
- Sustained release carrier

How to use

CSM-25/50



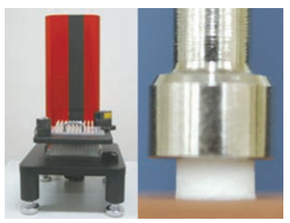
Cell proliferation test using Mighty (Internal data)



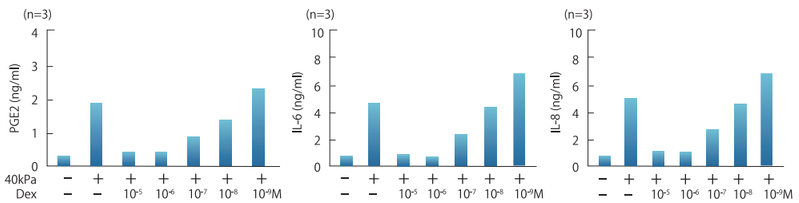
*Adding collagen to the cell suspension facilitates uniform cell seeding and uniform pressurization and prevents Atelcollagen Sponge MIGHTY from collapsing due to excessive pressur.

Example 1 Mechanical stress culture experiment of synovium-derived cells using MIGHTY (*in vitro* arthritis model)

(Shimomura K, Nakata K, Osaka University Graduate School of Medicine, Health and Sports Science (Sports Medicine))



Overview of CLS [manufactured by Techno View Co., Ltd.](left).
Image of compressive load (right).



Amount of protein in culture supernatant after mechanical stimulation culture with addition of dexamethasone

Human synovium-derived cell suspension was mixed with atelocollagen and seeded onto MIGHTY. After culturing for 3 days, a cyclic loading of 40 kPa was applied for 1 hour using a cyclic load stimulator (CLS).
As a result, increased expression of PGE2, IL-6, and IL-8, which are involved in the onset of arthritis, was observed.
This shows that MIGHTY is useful as an *in vitro* arthritis model. (Reference 8)

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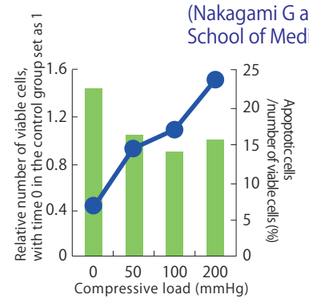
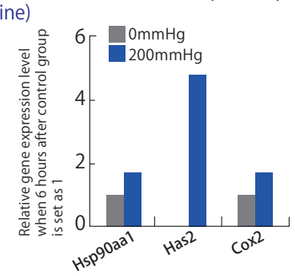
Example 2 Mechanical stress culture of fibroblasts using MIGHTY (*in vitro* granulation compression model)

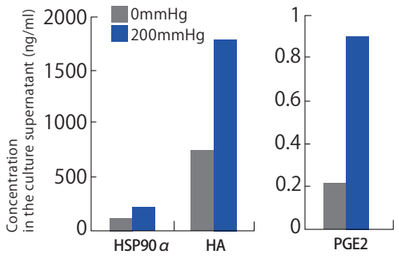
(Nakagami G and Sanada H, The University of Tokyo, Department of Geriatric Nursing, Graduate School of Medicine)



Relative viable cell number and apoptosis cell number rate after 6 hours of mechanical stress culture



Relative expression level of each gene after 6 hours of mechanical stress culture

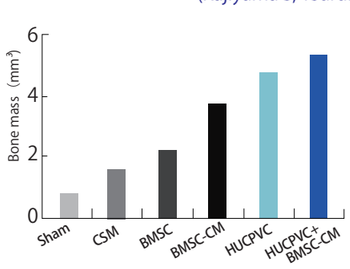


Concentration of each molecule in the culture supernatant after 6 hours of mechanical stress culture at 200 mmHg

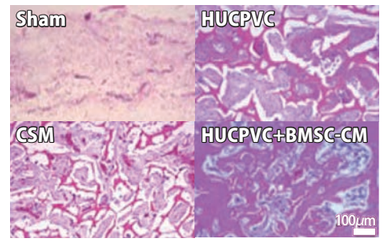
Rat fibroblast suspension was mixed with atelocollagen and seeded into MIGHTY. 1 day after culture, mechanical stimulation culture was performed using a unique compressive loading device. As a result, apoptosis increased in the cell group subjected to mechanical stress culture. In addition, the expression levels of HSP90 α, hyaluronic acid (HA), and COX-2 genes increased. HSP90a acts to repair damaged proteins and promote cell survival; HA is involved in the load resistance of the extracellular matrix; and COX-2 is a gene whose expression is enhanced by interaction with HA and CD44. Furthermore, the concentrations of HSP90 α, HA, and PGE2, a downstream molecule of COX-2, in the culture supernatant increased with mechanical stress culture. This suggests that these genes may be biomarkers for delayed pressure injury healing. (Reference 7)

Example 3 Transplantation of umbilical cord perivascular cells into cranium defects and osteogenesis using Mighty

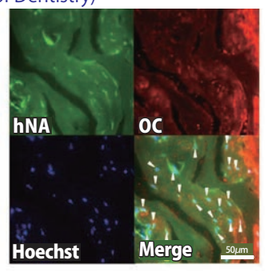
(Kajiyama S, Tsurumi University, Department of Periodontology, Faculty of Dentistry)



Bone mass evaluation 4 weeks after transplantation



H&E staining image 4 weeks after transplantation

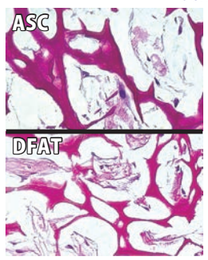


Fluorescence microscopic image in the HUCPVC+BMSC-CM group 4 weeks after transplantation

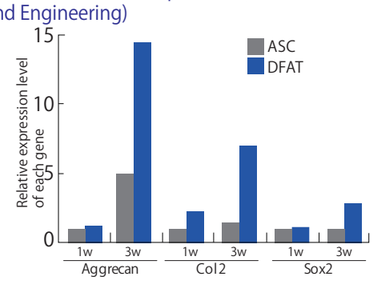
Human bone marrow-derived mesenchymal stem cells (BMSCs), human BMSC conditioned medium (BMSC-CM), human umbilical cord pericytes (HUCPVC), and HUCPVC+BMSC-CM were seeded or added to Mighty and transplanted into rat cranium defects. As a result, although no significant difference was observed between the HUCPVC group and the HUCPVC+BMSC-CM group, both groups showed a significant increase in osteogenesis compared to the Sham group and the CSM alone transplant group. H&E staining revealed that soft tissue was conspicuous in the CSM-only group, whereas neonatal bone growth into the CSM was observed in the HUCPVC and HUCPVC+BMSC-CM groups. And survival and osteogenic differentiation of transplanted HUCPVC were observed by immunofluorescence staining. (Reference 2)

Example 4 Induction of cartilage differentiation of dedifferentiated fat cell and adipose-derived regenerative cells using Mighty

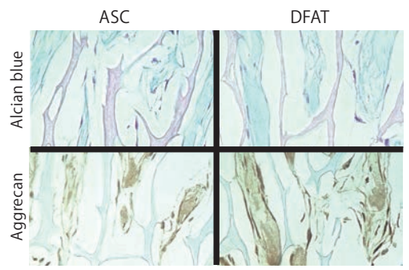
(Nishio N, Osaka Dental University, Second Department of Oral Surgery, Faculty of Dentistry, Osaka Dental University (at the time of data provision) / Hashimoto Y, Osaka Dental University, Department of Dental Science and Engineering)



H&E staining image of central section of Mighty seeded with ASC and DFAT



Expression levels of each gene 1 week and 3 weeks after induction of chondrogenic differentiation of ASC and DFAT



Alcian blue staining and Aggrecan immunostaining images after 3 weeks of ASC and DFAT cartilage differentiation induction

When a suspension of Human buccal fat pad adipose-derived stem cells (ASC) or dedifferentiated fat cells (DFAT)was mixed with atelocollagen and seeded on Mighty, uniform cell seeding was observed not only in the upper and lower parts of Mighty but also in the center. When these cells were cultured in cartilage differentiation medium for 3 weeks, the expression levels of cartilage cell markers Aggrecan, Collagen Type 2 (Col2), and Sox2 were significantly increased in the DFAT group compared to the ASC group. (Reference 3)