Xyltech™ H-Fbro-01

Catalog Number (BBARL/NIPRO): 10301/87-281 100 mL

1. Product features

This product is a synthetic culture medium suitable for suppressive growth control of normal human fibroblasts. Xyltech™ H-Fbro-01 can be used to control the growth rate of human fibroblasts in combination with Xyltech™ Growth H-Fbro, which is a serum-free medium for human fibroblast proliferation. This product is serum-free and contains no human or other animal-derived ingredients. If necessary, human serum (HS), fetal bovine serum (FBS), or serum substitute reagents can be added for cell culture.

2. Precautions for use

Xyltech™ H-Fbro-01 does not contain substances that neutralize trypsin activity. When subculturing cells with trypsin, it is strongly recommended that the trypsin activity be sufficiently neutralized with a trypsin inhibitor. Dilution washing alone does not completely remove trypsin activity and the remaining protease activity will reduce subsequent cell growth. This product is a research reagent. It cannot be used for human or animal treatment or diagnostic purposes.

3. Storage

Store Xyltech™ H-Fbro-01 in a cool, dark place (2-8°C). Do not freeze the medium to avoid deterioration of some active ingredients.

4. Example of suppressive growth control protocol for normal human fibroblast culture using Xyltech™ Growth H-Fbro-01

4-1. Cells and reagents

・Normal human fibroblasts (100 mm-dish)	
・Xyltech™ H-Fbro-01 (growth suppression medium)	***This product**
・Xyltech™ Growth H-Fbro (growth medium)	(CAT. # (BBARL/NIPRO): 10311/87-281)
・Artificial serum (Xf) or Artificial serum (Af)	(NIPRO CAT. #: 87-081/87-082)
・r-TE (r-Trypsin/EDTA Solution)	(NIPRO CAT. #: 87-974)
・s-TI (Synthetic Trypsin Inhibitor Solution)	(NIPRO CAT. #: 87-975)
・D-PBS(-)	

*We recommend adding 1% artificial serum (Xf) or artificial serum (Af) for use with Xyltech™ H-Fbro-01 and Xyltech™ Growth H-Fbro.

4-2. Growth control of normal human fibroblasts

1. Warm the culture medium, D-PBS (-), r-TE, and s-TI in a 37°C water bath.
2. Remove the culture supernatant of selected normal human fibroblasts that have reached around 80% confluence (sub confluence).
3. Rinse the cell layer with 5 mL of D-PBS (-).
4. Add 0.5 mL of r-TE and incubate at 37°C for approximately 2 minutes.
5. Add 0.5 mL of s-TI, mix well, gently pipette up and down several times, collect cells from the dish, and centrifuge for 1,000 rpm, 5 minutes.
6. Aspirate the supernatant and add the appropriate amount of Xyltech™ Growth H-Fbro medium to resuspend the cells and seed into a new tissue culture dish.
7. The next day, use a phase-contrast microscope to confirm the cells are engrafted and replace the growth medium with the Xyltech™ H-Fbro-01 culture medium to reduce the fibroblast growth rate.
8. The cells can be cultured for 3 days to 1 week with Xyltech™ H-Fbro-01. (The cells start to regrow quickly and become confluent 1-2 days after changing the medium back to the Xyltech™ Growth H-Fbro medium). Begin subculture and/or experiments with the cells.

4-3. Phase contrast microscope images of normal human fibroblasts cultured with Xyltech™ Growth H-Fbro or Xyltech™ H-Fbro-01

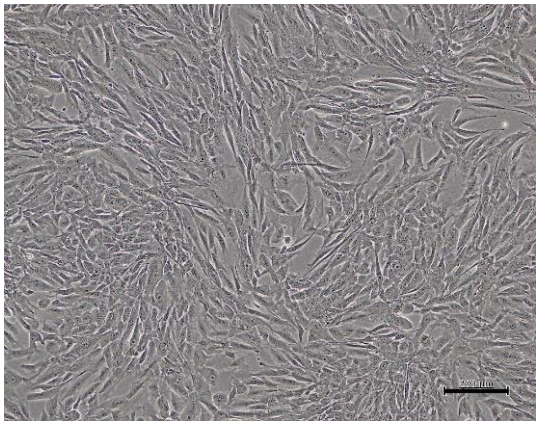
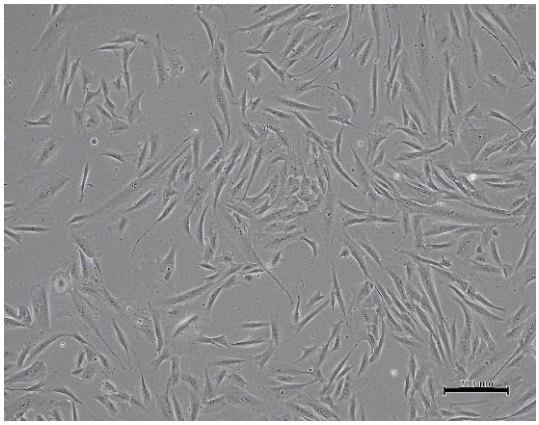


Fig. 1 Normal human fibroblasts cultured with Xyltech™ Growth H-Fbro for 3 days.



Bars=200 μm

Fig. 2 Normal human fibroblasts cultured with Xyltech™ H-Fbro-01 (growth suppressive medium) for 3 days.

*The protocol is based on experimental results. It may be necessary to adjust seeding density, and passage timing according to the cells. This protocol is intended for research purposes only.

5. For Inquiries about products

Bourbon Biomedical Advanced Research Laboratories, Inc. (BBARL, Inc.)
1-3-1, Ekimae, Kashiwazaki-city, Niigata,
TEL: +81-257-23-2769 E-mail: support@bourbon-barl.co.jp