

# Biofilm-Clearing Reagent iCBiofilm

Clearing Reagent iCBiofilm-H1 [for Biofilm]

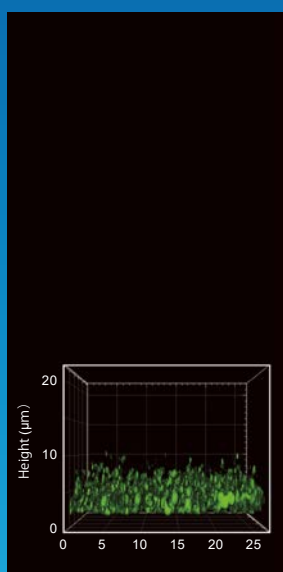
8mL [T4031]

Clearing Reagent iCBiofilm-H2 [for Biofilm]

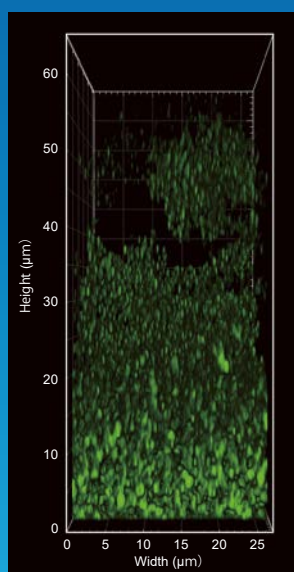
8mL [T4032]

Clearing Reagent iCBiofilm-D [for Biofilm]

8mL [I1275]



PBS



iCBiofilm-H1

## Advantages

- Makes biofilm transparent by simply adding iCBiofilm.
- Suitable for various kinds of biofilm.
- Cleared biofilm can be observed by light-sheet fluorescent microscopy (LSFM) or confocal laser-scanning microscopy (CLSM).
- iCBiofilm-D [I1275] allows for live cell imaging of biofilms.

Images of methicillin-resistant *Staphylococcus aureus* (MRSA) observed microscopically after incubated overnight at 37°C, fixed at room temperature for 30 minutes, stained with FM<sup>®</sup>1-43 at 4°C overnight, washed, and dipped in PBS or iCBiofilm-H1. (FM<sup>®</sup> is a trademark of Thermo Fisher Scientific Inc.)

## Application

iCBiofilm-H1 [T4031] is applicable to clearing many kinds of biofilms. If the transparency with iCBiofilm-H1 is insufficient, iCBiofilm-H2 [T4032] can be used instead. Since iCBiofilm-H2 is prepared at high component concentration, iCBiofilm-H2 is available as a stock solution. Before addition to biofilm, iCBiofilm-H2 has to be diluted with deionized water to the appropriate concentration. (Undiluted iCBiofilm-H2 may also work well).

### [Direction for Use]

1. Prepare a biofilm in a glass bottomed culture dish (35mm diam.).
2. After removing planktonic cells and the culture medium from the dish, add 1% glutaraldehyde solution diluted with PBS to fix the biofilm at room temperature for 30 minutes. To preserve immunogenicity for immunofluorescence applications, the use of 4% paraformaldehyde in PBS is recommended.
3. Remove the glutaraldehyde solution and wash three times with PBS or distilled water.
4. Stain the biofilm with appropriate fluorescence reagents.
5. Remove staining reagents and add iCBiofilm solution.
6. Observe the biofilm by optical microscopy.

These products are commercialized under the instruction of Assoc. Prof. Shinya Sugimoto.

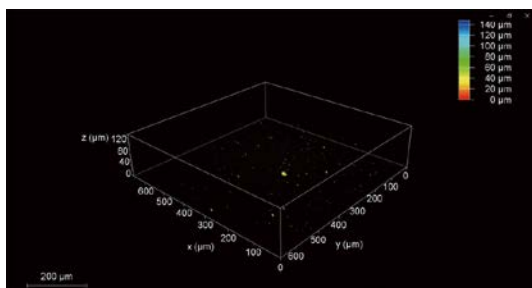
# Biofilm-Clearing Reagent: iCBiofilm

## Application: Live-Cell-Imaging of Biofilms

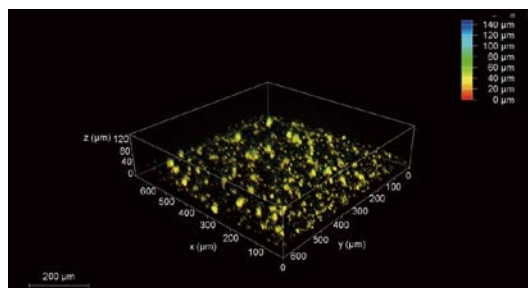
### [Directions for Use]

1. Prepare live-cell-imaging medium:  
Dissolve [1] Brain Heart Infusion (BHI) powder to twice the normal culture concentration and [2] glucose powder to a final concentration of 2% in deionized water and filter sterilize. Then mix with an equal volume of iCBiofilm-D [I1275] to make BHI medium containing 1% glucose with 50% iCBiofilm-D.
2. Add MitoTracker® Deep Red to the live-cell-imaging medium and culture Methicillin-Resistant *Staphylococcus Aureus* (MRSA) in a glass bottom dish.
3. Observe the biofilm via microscopy to obtain time lapse images.

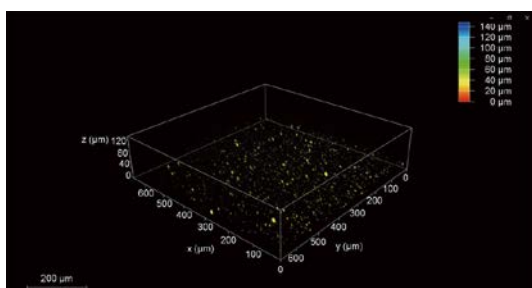
MitoTracker is a registered trademark of Thermo Fisher Scientific Inc.



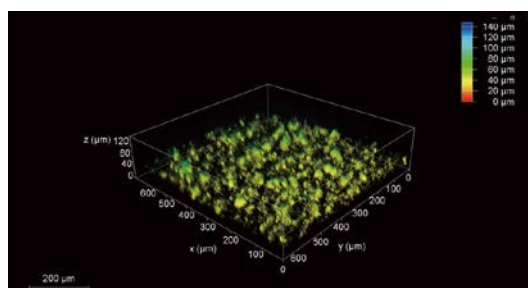
0 hour



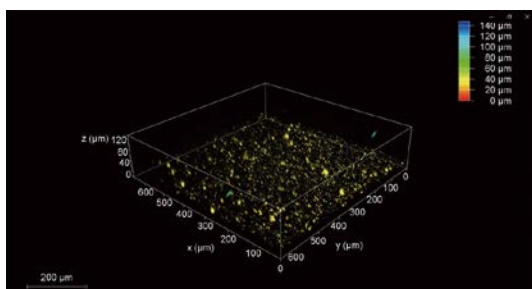
6 hours



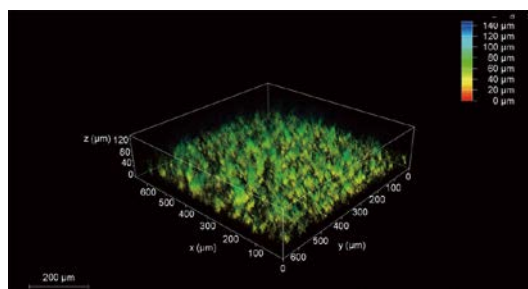
2 hours



12 hours



4 hours



16 hours

### Note:

If clear images cannot be obtained or biofilm formation is prevented when first trying 100% iCBiofilm-D, try diluting iCBiofilm-D with culture medium at a ratio of 1:1 according to the above application method. If iCBiofilm-D must be used at the original concentration, but with culture medium, prepare live-cell-imaging medium using iCBiofilm-D in place of water.

### Ordering and Customer Service

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