

**Product Name**  
Mouse Anti-Flavobacterium psychrophilum  
Fluorescein Isothiocyanate Conjugated Monoclonal  
Antibody

**CAT No.**  
CC0116F

**LOT No.**  
35967C

**Quantity**  
140µg

  
products@immunoprecise.com  
www.immunoprecise.com

#### Applications

Immunofluorescence (IF)  
See references for detailed applications of this  
antibody

#### Antibody Concentration / Working Dilution

0.73 mg/mL / use at 1/100 – 1/500

#### Source

Protein G purified from bioreactor concentrated  
supernatant

#### Host/Isotype/Clone ID

Mouse, IgG2b, FL43

#### Reconstitution

Add 140µL of deionized water to lyophilized  
sample to yield 1mg/mL solution

#### Background

There is strong evidence that *Flavobacterium psychrophilum*, the etiologic agent of coldwater disease (CWD), is transmitted vertically and it has been hypothesized that disease management at hatchery facilities may be improved through broodstock screening and implementation of culling programs. FL43 was selected for assay development and shown to react with 67 *F. 31 psychrophilum* isolates tested, but did not react with two strains of *Flavobacterium columnare* or 32 one strain each of *F. pectinovorum*, *F. aquatile*, *F. branchiophilum*, and *F. saccharophilum*.

#### Species Cross Reactivity / Specificity

MAb FL43 exhibited specificity to *F. psychrophilum* isolates CSF 259-93 and ATCC 49418. No cross reactivity was observed with *F. columnare* GA-02 or ATCC 23463.

#### Formulation

**Buffer:** 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

**Stabilizer:** 10 mg/mL Bovine Serum Albumin (BSA), IgG and Protease free

**Preservative:** 0.01% (w/v) Sodium Azide

#### Recommended Dilutions

Optimal titres for applications should be determined by the researcher. Range 1/100-1/500

#### Stability/storage

+4°C (do not freeze) stable 6 months after reconstitution.  
Dilute only prior to immediate use.

#### Immunogen

Outer membrane fractions (OMF) of *F. 116 psychrophilum* CSF 259-93 were prepared following the method of Filip et al. (1973)

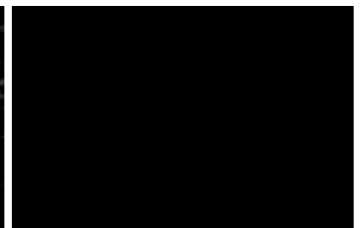
#### Sample Data - IF

*Flavobacterium psychrophilum* stained with MAb FL43 conjugated to AlexaFluor-488 and viewed on a glass slide. *F. columnare* and *F. pectinovorum* served as negative controls. The images were viewed with epifluorescent microscope housing a FITC filter. Photomicrographs are at 100x magnification.

*F. psychrophilum*



*F. columnare*



#### References

Lindstrom, N. M., Call, D. R., House, M. L., Moffitt, C. M., and Cain, K. D. A quantitative enzyme-linked immunosorbent assay (ELISA) and filtration-based 1 fluorescent antibody test (FAT) as potential tools to screen broodstock for 2 *Flavobacterium psychrophilum* infection. *Journal of Aquatic Animal Health* 2009; 21: 43-56

Filip, C., Fletcher, G., Wulff, J.L. & Earhart, C.F. (1973). Solubilisation of the cytoplasmic membrane of *Escherichia coli* by the ionic detergent sodium-lauryl sarcosinate. *Journal of Bacteriology* 115, 717-722.

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### **Fluorescent Antibody Test**

\* Bacteria should be grown in a broth culture and harvested during the exponential growth phase to ensure optimal results from this procedure.

1. Prepare a solution of PBS + 0.5% (w/v) non-fat dry milk (NFDM). You will need 100 µl per sample being processed.
2. Mix 100 µl of exponentially growing bacteria with 100 µl of PBS + NFDM in a 1.5 ml microcentrifuge tube.
3. Incubate the solution at room temperature for 15 minutes.
4. Dilute MAb FL-43:FITC 1:100 in PBS + NFDM. The amount of diluted antibody added to the sample will be equal to the total sample volume. For example, 200 µl of diluted antibody is mixed with 200 µl of sample (100 µl sample and 100 µl PBS).
5. Incubate the mixture in the dark at room temperature for 30 minutes.
6. Spin the culture at 3500 xg for 10 minutes.
7. Remove the supernatant and wash the pellet in an equal volume of PBS.
8. Repeat the spin and washing of the pellet 2 times. After the last spin, remove the supernatant and resuspend the pellet in 100 µl PBS.
9. Pipet 10 µl of the solution on to a slide and place a coverslip over it.
10. Slide should be analyzed immediately with an epifluorescence microscope equipped with a FITC filter.