

# Detect cholinesterase.

Use Ph-F to label cells containing active cholinesterase.


## WHAT IS PH-F?

With ICT's cholinesterase assay (Ph-F, patent pending), researchers now have an easy way to directly quantitate and localize active cholinesterase enzymes in live, intact cells - no lysis or permeabilization steps are required. This unique kit is not an ELISA and does not use antibodies - instead it is based on physostigmine, a known cholinesterase inhibitor, linked to a green fluorescent label, fluorescein (F).

ICT's Ph-F reagent is cell permeant, so you don't have to lyse the cells or permeabilize the membranes. Because this kit does not use antibodies, there is no cross-reaction with inactive or pro forms of the enzyme. Only cells with the active cholinesterase enzyme will fluoresce.

Simply culture your cells, add the cholinesterase reagent to the media, incubate, and wash the cells. If active cholinesterase enzymes are present, they will bind to Ph-F, and retain it on or within the cell, generating a green fluorescent signal. Analyze samples with a fluorescence microscope, plate reader, or a flow cytometer. Green Ph-F excites at 490 and emits at >520nm. Trial size kits contain enough reagent for ~25 tests; regular size kits contain enough reagent for ~100 tests.

## *in vitro* cholinesterase

 **Cholinesterase, green detection probe**  
reagent, fixative, and wash buffer

Physostigmine-Fluorescein  #973, trial size  
 #974, regular size

for more info go to [www.immunochemistry.com/Cholinesterase.htm](http://www.immunochemistry.com/Cholinesterase.htm)

## CHOLINESTERASE



(A) DAPI (B) Ph-F (C) SR-VAD-FMK

Figure 1: Varying expressions of cellular DNA (A), cholinesterase (B), and caspase activity (C) in apoptotic Jurkat cells.

Jurkat cells were treated with 20  $\mu$ M Ph-F (B), 10  $\mu$ M SR-VAD-FMK (C) for 1 hour, and counterstained with 1  $\mu$ g/mL of DAPI (A). The cells were viewed using UV- (A), blue- (B) or green- (C) incident light illumination to excite DAPI, Ph-F or SR-VAD-FMK (FLICA™), respectively. Cell #1 has active cholinesterase (it is green, B), but has no caspase activity (it is not red in C), nor does it reveal cellular DNA (it is not blue in A). Cells #2 and #3 reveal cellular DNA and cholinesterase and caspase activity at varied intensities. Data courtesy of X. Huang, Brander Cancer Inst., NY Medical College, NY).



"TRY a CHOLINESTERASE  
KIT TODAY!  
CALL ICT AT 800-829-3194!"

## CHOLINESTERASE

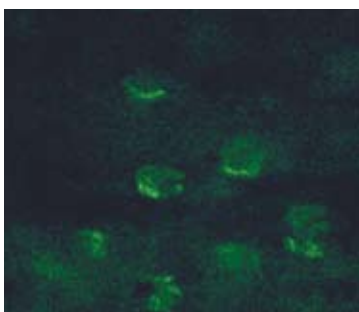


Figure 2: Localization of cholinesterase with Ph-F in the nerve-muscle junctions (end-plates) in C57 mouse diaphragm muscle tissue. The diaphragm muscle was dissected and fragments of it incubated in PBS containing 20  $\mu$ M Ph-F for 1 hour. The muscle was then rinsed for 20 minutes in PBS, stretched on a microscope slide, mounted under a coverslip, and examined by fluorescence microscopy using blue light excitation. Notice the retention of the fluorochrome.