Phosphatases are a specific class of enzymes that catalyze the removal of phosphate groups from proteins, whereas kinases enzymatically add a phosphate to a protein. Phosphatases are important in signal transduction because they regulate the proteins they are attached to. To reverse the regulatory effect, the phosphate has to be removed. Cell proliferation and differentiation are regulated by phosphatases.

Phosphatases serve also as enzyme markers, allowing to quantify phosphatase activity in different types of cells. Alkaline phosphatase is finally a highly sensitive enzyme for ELISA, immuno-histochemical, Northern, Southern and Western blot applications.

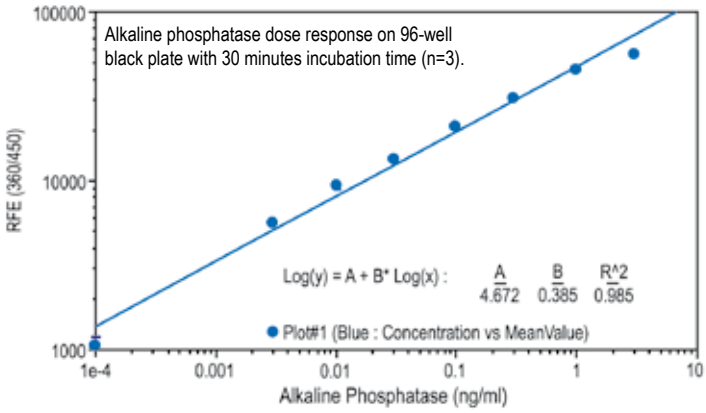
■ Fluorimetric Assay kits

The Alkaline Phosphatase Assay Kits use fluorogenic phosphatase substrate, to quantify alkaline phosphatase activity in solutions, in cell extracts, in live cells as well as on solid surfaces (such as PVDF membranes). The kits provide all the essential components with our optimized "mix and read" assay protocol that is compatible with HTS liquid handling instruments.

■ Alkaline Phosphatase Assay Kit, Blue Fluorescence

Substrate : MUP
$\lambda_{ex./em.}$: 360 / 449 nm
Sensitivity : 0.3 pg of alkaline phosphatase

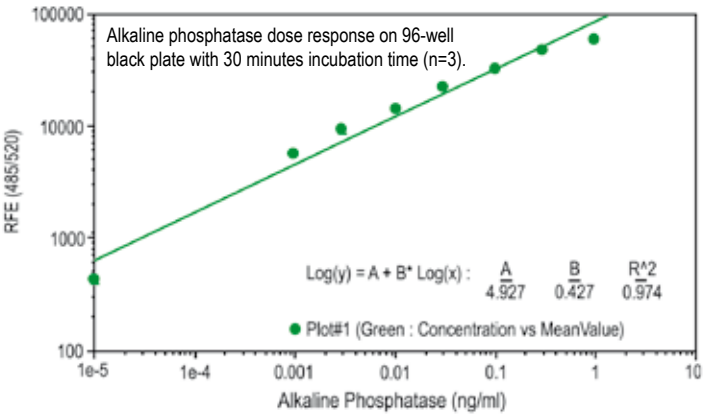
Description	P/N :	Qty
Fluorimetric Alkaline Phosphatase Assay Kit, Blue fluorescence	**JQ6730**	500 tests



■ Alkaline Phosphatase Assay Kit, Green Fluorescence

Substrate : FDP.
$\lambda_{ex./em.}$: 490 / 514 nm
Sensitivity : 0.1 pg of alkaline phosphatase

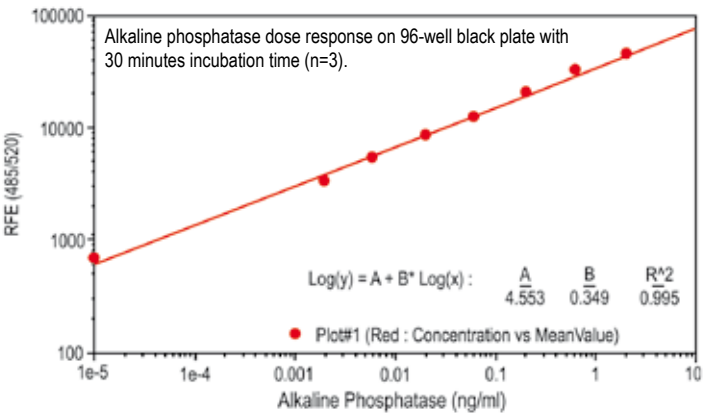
Description	P/N :	Qty
Fluorimetric Alkaline Phosphatase Assay Kit, Green fluorescence	**JQ6740**	500 tests



■ Alkaline Phosphatase Assay Kit, Red Fluorescence

Substrate : Phospholite™ Red
$\lambda_{ex./em.}$: 570 / 590 nm
Sensitivity : 0.2 pg of alkaline phosphatase

Description	P/N :	Qty
Fluorimetric Alkaline Phosphatase Assay Kit, Red fluorescence	**JQ6750**	500 tests

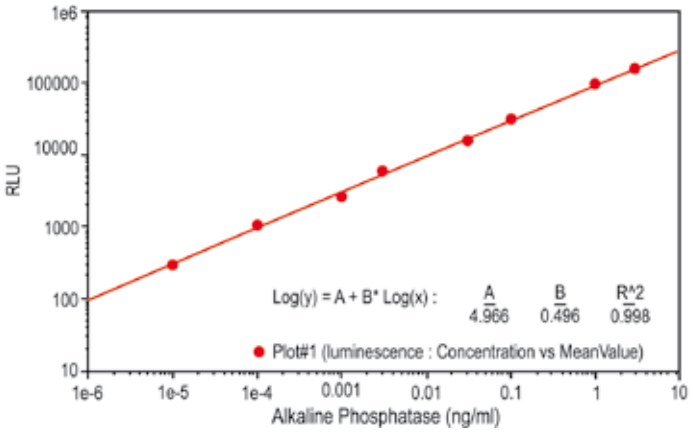


Luminometric AP Assay kit

■ Luminometric Alkaline Phosphatase Assay Kit

Substrate : proprietary luminescent substrate
$\lambda_{em.}$: **560 nm**
Sensitivity : **0.01 pg** of alkaline phosphatase

This Alkaline Phosphatase Assay Kit uses a luminogenic phosphatase substrate, to quantify alkaline phosphatase activity in solutions, in cell extracts, in live cells as well as on solid surfaces (such as PVDF membranes). This proprietary phosphatase substrate generates a luminescent product that produces strong luminescence upon interaction with phosphatase.



Alkaline phosphatase dose response on 96-well black plate with 30 minutes incubation time (n=3).

Description	P/N :	Qty
Luminometric Alkaline Phosphatase Assay Kit	**JQ6760**	200 assays

Colorimetric AP Assay kits

See AP colorimetric assays substrates and kits in Immunoassay reagents section.

Acide phosphatase assays

■ MUP Plus

Although MUP is widely used for detecting phosphatases in solution it is not well suited for living cell or continuous assays since 4-methylumbelliferone, the enzymatic product, which only develops maximum fluorescence at pH value of >10. Thus it is also difficult to use MUP for the detection of phosphatases that have acidic optimal pH range such as acid phosphatases. FluoProbes is pleased to offer MUP Plus that is developed to address this pH limitation associated with MUP substrates. MUP exhibits maximum fluorescence above pH 7.0, thus MUP Plus substrate can be well used for continuous assays. It can also be used for the assays that require acidic pH such as acid phosphatases.

Description	P/N :	Qty
MUP Plus™, sodium salt $\lambda_{ex./em.}$: 360 / 450 nm MW : 300 g.mol^{-1}	**FP-JQ6710**	25 mg

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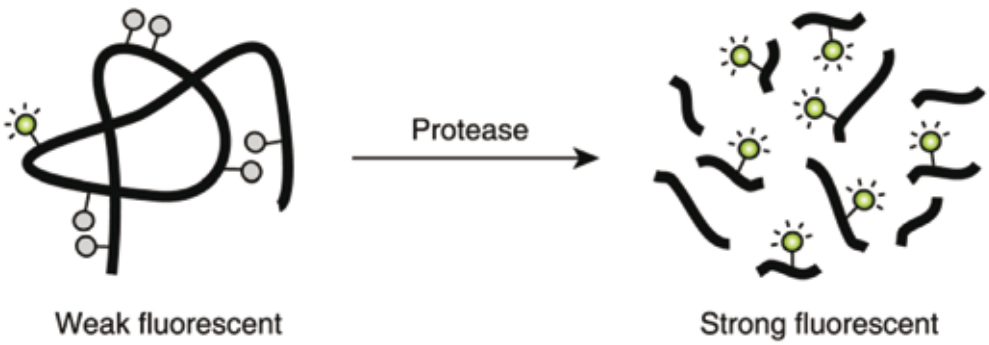
Protease Assays Kits, Green & Red Fluorescence

- Optimized Performance : Optimal conditions for the detection of **generic protease activity**
- **High Speed** : Minimal hands-on time
- **Assured Reliability** : Detailed protocol and references are provided

The Protease Assay Kits are widely used for detection of generic protease activities. The kits use a casein derivative that is heavily labeled with green or red fluorescence, resulting in almost total quenching of the conjugate's fluorescence. Protease-catalyzed hydrolysis relieves this quenching conjugate, yielding brightly fluorescent dye-labeled peptides. The increase in fluorescence intensity is directly proportional to protease activity. The protease assay kits do not require any separation steps and can be used to continuously measure the kinetics of a variety of exopeptidases and endopeptidases.

The kits contains :
• Fluorescent labeled casein with high ratio of dye/protein (pH-insensitive)
• Trypsin (as positive control)
• Assay buffer
• A detailed protocol

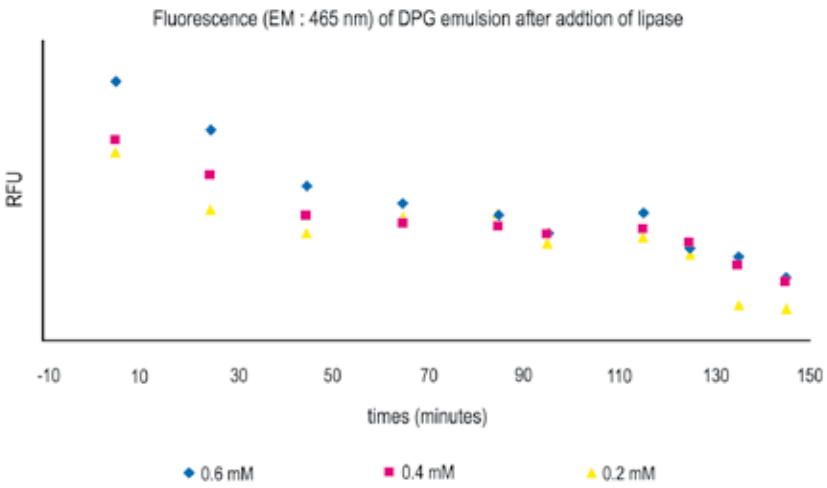


Description	P/N :	Qty
Protease Assay Kit, Green Fluorescence (488/520 nm)	**BK962A**	500 tests*
Protease Assay Kit, Red Fluorescence (546/575 nm)	**BK963A**	500 tests*

Lipase Assay Kit

Fast and easy measurement of lipase activity in vitro, in cell preparations or in vivo

Lipases are a family of enzymes that release fatty acids from triacylglycerols in a site specific manner. Most lipases have optimum activity for the primary ester groups of triglycerides, while some lipases remove fatty acyl groups from either the C-1 or C-3 acyl positions. The substrate is typically not a single molecule, but a nonaqueous phase of aggregated lipid. Lipase activity, ubiquitous among most cells, can be monitored using the new fluorescent substrate 1,2-dioleoyl-3-pyrenyldecanoyl-rac-glycerol (Product # FP-M14031, $\lambda_{exc./em.}$: 342/470 nm) contained in the kit. Upon cleavage, the fluorescent fatty acid pyrenedecanoic acid (Product # FP-37853A, $\lambda_{exc./em.}$: 341/377 nm) is released and activity measurements are easily obtained either in vitro, in cell preparations, or in vivo. The kit contains enough substrate for numerous assays and control experiments, and also contains reference standards and a detailed protocol for use.

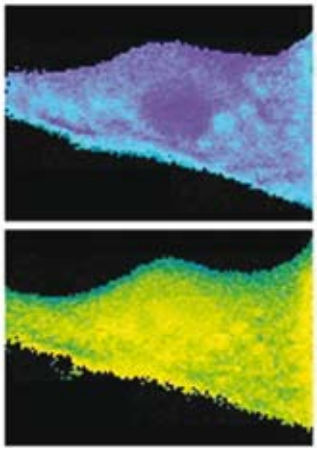


References :
Howard G.T., et al. "Sensitive plate assay for screening and detection of bacterial polyurethanase activity". *Lett. Appl. Microbiol.* 32(3): 211-4 (2001)
Kouker G. and Jaeger K.E., "Specific and sensitive plate assay for bacterial lipases". *Appl Environ Microbiol* 53(1): 211-3 (1987)

Description	P/N :	Qty
Fluorescent Lipase Assay Kit	**BG8440**	1 kit (72 assays in 96 well plate)

Unbound free fatty acids Assay - ADIFAB

The assay can be used in a variety of biochemical and clinical applications including the determination of lipase activity, fatty acid binding to membranes and proteins, and serum unbound free fatty acid (FFAu) levels.
ADIFAB and ADIFAB2 are ideal also for drug screening. They are particularly well-suited for drugs that affect cellular processes as well as those involving purified enzymes.
FFAu probes have been validated for high throughput assay. This includes robotic dispensing of reagents, fluorescence screening and the determination of FFAu levels in 96 and 384 well plates. This system can be used directly to screen for drugs that alter cellular metabolism involving FFA or that alter the behavior of enzymes that either produce or use FFA.
ADIFAB2 is a high affinity version of the original ADIFAB probe. It is formed by acrylodan labeling the Leu72 to Ala mutant of the Intestinal Fatty Acid Binding Protein. ADIFAB2, similarly to ADIFAB, can be used to assay unbound free fatty acid levels but provides greater sensitivity for low concentrations of the FFA levels. For those concentrations below about 400 nM, the increase in the ADIFAB2 emission ratio is about twice that for ADIFAB.
The fluorescence of ADIFAB is measured with the ratio 505/432 upon excitation at 386 nm. ADIFAB2 fluorescence emission spectra occur at longer wavelengths (550/457 nm with excitation at 375 nm). Binding affinities for ADIFAB2 are approximately ten times greater than for ADIFAB. On the other hand, ADIFAB has a wider range of sensitivity than ADIFAB2 and is preferable for higher concentrations of unbound free fatty acids.



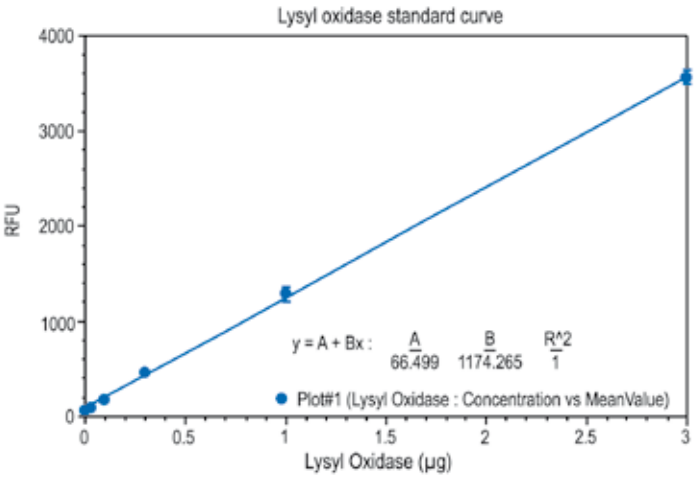
Intracellular FFAu levels measured with ADIFAB micro-injected into fat cells – Copyright FFA biosciences

Description	P/N :	Qty
ADIFAB	**040791**	200 µg
	0470792	1 mg
ADIFAB2	**BB6681**	200 µg
	BB6682	1 mg

■ Lysyl Oxidase Assay Kit, Red Fluorescence

Sensitivity : **40 ng** of lysyl oxidase in solution

Lysyl oxidase is an extracellular enzyme that catalyzes formation of aldehydes from lysine residues in collagen and elastin precursors. These aldehydes are highly reactive, and undergo spontaneous chemical reactions with other lysyl oxidase-derived aldehyde residues, or with unmodified lysine residues. This results in cross-linking collagen and elastin which is essential for stabilization of collagen fibrils and for the integrity and elasticity of mature elastin. Lysyl oxidase has been identified as a possible tumor suppressor. Lysyl oxidase activity in biological samples is traditionally and most reliably assessed by tritium release end-point assays using radiolabeled collagen or elastin substrates involving laborious vacuum distillation of the released tritiated water. This kit offers a sensitive fluorescent assay for lysyl oxidase activity that utilizes 1,5-diaminopentane as substrate, and released hydrogen peroxide is detected using our HRP substrate in HRP-coupled reactions. This method allows the detection of sub ng/mL lysyl oxidase and is much more sensitive than the currently available fluorimetric assay for this enzyme activity. This method eliminates the interference that occurs in some biological samples and can be readily used to detect lysyl oxidase activity in cell culture experiments.



Lysyl oxidase dose response on 96-well black plate with 30 minutes incubation time (n=3). The insert shows the low levels of lysyl oxidase detection.

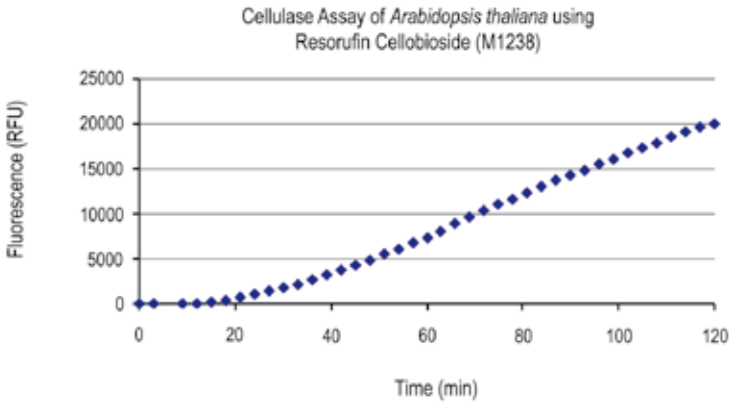
Description	P/N :	Qty
Lysyl Oxidase Assay Kit, red fluorescence	**JQ7270**	500 assays

Cellulase Detection

■ Cellulase Assay Kit, Red Fluorescence

Substrate : Resorufin Cellobioside
Reaction volume : 100 µl
$\lambda_{ex./em.}$: **571/585 nm**

Cellulases are a family of enzymes that include ß-Glucosidases, endoglucanases, and exoglucanases. These enzymes cleave the ß-1,4-D-glycosidic bonds that link the glucose units comprising cellulose. In addition to being produced by plants, cellulase activity is found in many fungi and bacteria, including some plant pathogens. Most animal cells are not known to produce cellulase; cellulolytic activity is often carried out in animals by symbionts. However, recent evidence does suggest cellulase production in some animals, such as insects and arthopods. The study of cellulase activity has many applications in plant molecular biology, agriculture, and manufacturing.



Suspension of flowering buds from two mature *Arabidopsis thaliana* plants in triplicate (50 µL) on a 96-well clear, flat bottom plate read at 3-minute intervals for 120 minutes.

Cellulase is also becoming important in the development of alternative fuel sources, as glucose obtained from cellulose hydrolysis is easily fermented into ethanol. Activity of most cellulases can be monitored using our long wavelength fluorescent substrate, Resorufin Cellobioside, contained in the kit. Upon cleavage, the fluorescent compound, Resorufin, is released and activity measurements are easily obtained in a microtiter plate based assay format.

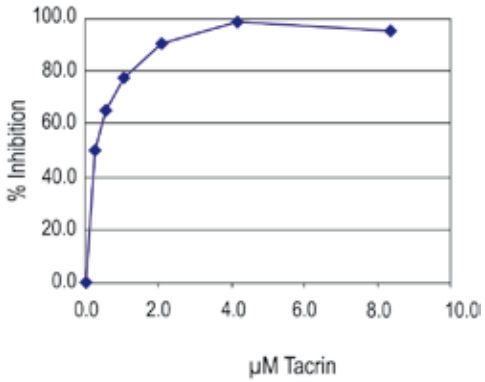
Description	P/N :	Qty
Fluorescent Cellulase Assay Kit	**DO8110**	200 assays
Kit contains : Substrate Reagent, Reference Standard, Reaction Buffer, Stop Buffer, DMSO		

aCella – Acetylcholinesterase Assay

Bioluminescence assay for Monitoring AcetylCholinEsterase Activity

- FAST : Results in **30 seconds - 5 minutes**
- Homogenous : **One-step, no wash assay**
- **Ultra Sensitive** assay to monitor AChE activity
- **Versatile** : Nerve gas, pesticide monitoring ; drug screening applications

Acetylcholinesterase (AChE) is one of the most important enzymes involved in nerve transmission. The enzyme is bound to cellular membranes of excitable tissue (synaptic junction, endoplasmic reticulum, etc). Acute toxicity to humans and animals through inhibition of AChE by both nerve gases and an important class of pesticides has long been a field of intensive scientific investigation. AChE inhibitors have also been used clinically as Alzheimer's treatments (e.g., tacrine (tetrahydroaminoacridine)) and are the subject of increasing interest in various disease processes and treatment strategies. However, both environmental detection of AChE inhibitors and development of modulators of AChE enzymatic activity as drugs have been hampered by the difficulty and complexity of the current assay methods.



Tacrine (a mixed-mode inhibitor of AChE) was serially diluted in DI water. Next 10 µL of the diluted Tacrine (x axis labeling represents µM final concentration of Tacrine) was added to a white opaque 96 well microplate along with 50 µL of component A (AChE enzyme). The samples were incubated for 5 minutes after which 50 µL of component B was added to all the wells. Data was collected using a luminometer. Data shown represents T=2 minutes after the addition of component B.

Assay Principle

Reaction I : AChE + Inhibitor → No Acetate and Choline.

Reaction II : Coupled enzyme reaction + ATP → Reaction does not proceed.

Reaction III : ATP (remaining) + Luciferase/Luciferin → LIGHT

Description	P/N :	Qty
aCella –AchE Assay	**CA6650**	100 tests
	CA6651	500 tests
	CA6652	1000 tests

Kit Contents : Acetylcholinesterase, Detection reagent, acetylcholine and coupled enzyme reaction, Control to measure maximum luminescence

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