

Gelatin-Fluorescein

Product Description

Substrate of gelatinases and collagenases.

Name :	Gelatin - Fluorescein conjugate Gelatin extracted from pig skin, conjugated to 5-fluorescein
Catalog Number :	FP-M1959A, 5mg
Solubility:	soluble in water
Fluorescence:	$\lambda_{exc} \mid \lambda_{em} = 492/515 nm$

Storage: -20°C desiccated and protected from light ^(M) Protect from light and moisture

Directions for use

Gelatin is denatured collagen composed of heterogeneous mixture of water-soluble proteins of high average molecular weights. Below 35-40°C gelatin swells and absorbs 5-10 times its weight of water to form a gel. Gel strength and viscosity gradually weaken upon prolonged heating in solution above 40°C. **Collagen** is a major component of the extracellular matrix (~25% of total protein in vertebrates). It not only serves a structural role, but is also important in cell adhesion and migration. Collagen and it's denatured form gelatin bind to specific collagen receptors, fibronectin and a number of other proteins involved in cell–cell and cell–surface adhesion.

Gelatin is extracted from porcine skin, and then heavily labeled with FITC.

Fluorescein-Gelatin can be used as a highly quenched substrate for the study of gelatinases and collagenases (metalloproteins that digest collagen and gelatin), but also for collagen-binding proteins and collagen metabolism. It is efficiently digested by gelatinases and collagenases, releasing brightly fluorescent peptides. The increase in fluorescence upon digestion is proportional to proteolytic activity. Longer incubation may increase its sensitivity for detecting proteases.

Directions for Use

The stock solution can be prepared by dissolving the product in distilled water (dH₂O) or PBS, to give 1 mg/mL solutions. The gelatin substrates may require sonication and heating to 50°C to aid dissolution. 10 mM acetic acid with heating to 50°C may also facilitate dissolution. Store solutions at 4°C with the addition of sodium azide at a final concentration of 2 mM. For long-term storage, divide solutions into aliquots and freeze at -20°C.

For gelatinolytic assays, dilute the stock solution in appropriate assay buffer, pH >7.0. Then incubate the Fluorescein-Gelatin with proteases at 37°C for 16-24 hr. Centrifuge the sample at 10 000 x g for 10 min. The protease activity is demonstrated by the increment of fluorescence in the supernatant at Ex/Em=490 nm/520 nm.

References

Otsuka, K. et al. (1997). An improved assay method for fibroblast gelatinolytic enzyme. J. Nihon. Univ. Sch. Dent. 39, 182-90.

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FT-M1959A

Related / associated products and documents

See BioSciences Innovations catalogue and e-search tool.

- Universal MMP activity assay kit, Green fluorescence, <u>CP1970</u>
- Universal MMP activity assay kit, Red fluorescence, <u>CP6030</u>

Ordering information

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<u>Catalog size quantities and prices may be found at www.interchim.com/</u> Please inquire for higher quantities (availability, shipment conditions).

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