FluoProbes[®]



FSB & other amyloid markers

beta amyloid plaques markers

Product Information

Product name cat.number	MW (g·mol ⁻¹)	λ _{exc} \λ _{em} . max. (nm)	Soluble in
FBS 1% solution FP-CG2370, 100 μl	420.39	390/511 nm	1% DMSO solution
BSB FP-BS6470, 5mg	481.3	340/520 nm	DMSO
BTA-1 FP-BS6480 10 mg	240.3	350/NA nm	DMSO
BTA-2 FP-BS6490 100 mg	268.4	356/437 nm	DMSO
Chrysamine G FP-BS6500 10 mg	482.5	386/NA nm	DMSO
Half Chrysamine G FP-BS6520, 10 mg	242.2	342/NA nm	DMSO
Congo Red N12511, 1g FP-AQ3370,100tests	696.7	497/NA nm	Water
ThioFlavin T FP-BS6530, 1g	318.9	412/482nm	DMSO

Storage: 0-5 °C, protect from light; Powders: dessicated.

Introduction

The formation of amyloid plaques and neurofibrillary tangles are thought to contribute to the degradation of the neurons (nerve cells) in the brain and the subsequent symptoms of Alzheimer's disease. One of the distinctive features of Alzheimer's disease is the accumulation of amyloid plaques between nerve cells (neurons) in the brain. These plaques consist primarily of the β -amyloid protein; however, other proteins, such as apoE, are also present.

In recent years there have been extensive studies to investigate the mechanism of Alzheimer's disease and the roles of β -amyloid in the cause of Alzheimer's disease. Interchim offers the comprehensive list of Amyloid markers, starting with the great FSB reagent (as well as (labeled) β -amyloid peptides, please inquire).



FT-CG2370

FSB

Name :	FSB , 1% DMSO solution [1-Fluoro-2,5-bis(3-carboxy-4-hydroxystyryl)benzene]
Catalog Number :	FP-CG2370 100 µl
Structure :	$C_{24}H_{17}FO_6$; MW= 420.39
Absorption / Emission :	$\lambda_{exc} = 390/511 \text{ nm}$

FSB, a bis-styrylbenzene analog with fluoride, stains beta amyloid plaques and emits strong fluorescence. The fluorescence intensity of FSB-Beta amyloid complex is twice as high as that of the BSB complex. FSB is also used for MRI detection of plaques due to fluoride in the structure. Higuchi, et al. demonstrated that this compound can be used to detect brain plaques of living mice.

Guidelines for use

- 1- Permeabilize 30 µm frozen sections of cortical regions
- 2- Block with 0.5% Triton X-100 and 5% goat serum in phosphate-buffered saline (PBS) for 20 minutes
- 3- Stain with primary antibodies
- 4- Wash with PBS/0.1% Triton X
- 5- Incubate the sections with FluoProbes conjugated secondary antibodies
- 6- Counterstain with a derivative of 10 μmol/L Congo red (*E*,*E*)-1-fluoro-2,5-bis(3-hydroxycarbonyl-4-hydroxy) styrylbenzene (FSB), which specifically binds to the β-sheet conformation of Aβ **plaques** with an excitation/emission wavelength of 390/511 nm.
- 7- Mount the sections on slides with Fluoromount G
- 8- Capture the confocal images of Aβ **plaque** regions using a confocal microscope with excitation at 488 (for **FSB**) laser.

Other protocols can be found in the literature.

Related products

- Fluoromount G mounting medium, <u>FP-483331</u>
- Triton X-100, <u>15851B</u>
- PBS 10X, <u>N14010</u> or PBS 20X, <u>N13760</u>
- Normal Goat serum, sterile, <u>UP379030</u>

- FluoProbes[®] 547H Goat anti-Mouse IgG, <u>FP-CB1020</u>
- FluoProbes[®] 547H Goat anti-Rabbit IgG, <u>FP-CB1050</u>
- FluoProbes[®] 647H Goat anti-Mouse IgG,<u>FP-CB1040</u>
- FluoProbes[®] 647H Goat anti-Rabbit, <u>FP-CB1060</u>

References

References:

- Bartlett S., MRI for in vivo detection of amyloid plaques, The Lancet Neurology, Volume 4, Issue 5, Pages 276-276 (2005)
- Higuchi M. et al., Nat. Neurosci., 8, 527-533 (2005)
- Small S., Alzheimer disease, in living color, Nature neuroscience, Vol. 8, 4:404-405 (2005) Article
- Sugiyama Y. et al., AFM and TEM observations of alpha-helix to β-sheet conformational change occurring on carbon nanotubes, Journal of Electron Microscopy 55(3):143-149 (2006) <u>Abstract</u>
- Yamamoto M. *et al.*, Interferon-gamma and Tumor Necrosis Factor-alpha Regulate Amyloid-β Plaque Deposition and β-Secretase Expression in Swedish Mutant APP Transgenic Mice, *American Journal of Pathology*. 170:680-692 (2007) <u>Article</u>

Other Amyloid markers

Name :	BSB [(trans,trans)-1-Bromo-2,5-bis-(3-hydroxycarbonyl-4-hydroxy)styrylbenzene]	
Catalog Number :	FP-BS6470 5 mg	
	MW: 481.3; Soluble in DMSO	
	$\lambda_{\rm exc} \lambda_{\rm em} = 340/520 \text{ nm}$	

BSB, derived from the structure of Congo Red, is shown to bind to a wide range of amyloid inclusions in situ. More importantly it is also used to label brain amyloids in live animals.

BSB recognizes amyloid lesions, and has distinctive properties which allow the quantitative monitoring of the formation of amyloid fibrils assembled from the Ab peptide, a-synuclein and tau. It is a cell-permeable fluorescent probe that specifically binds to and labels intracellular b-amyloid aggregates both in vitro (Ki = 400 nM) and in vivo. It is also used as an antemortem diagnostic tool for animal models of aalzeimer's disease.

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FT-CG2370

Crystal AS, et al. (2003). A comparison of amyloid fibrillogenesis using the novel fluorescent compound K114. J Neurochem 86, 1359-68. Kung MP, et al. (2002). Radioiodinated styrylbenzene derivatives as potential SPECT imaging agents for amyloid plaque detection in Alzheimer"s disease. J Mol Neurosci 19, 7-10.

Name :	BTA-1 [2-(4"-(methylamino)phenyl)benzothiazole]
Catalog Number :	FP-BS6480 10 mg
U U	MW: 240.3
	$\lambda_{exc} = 350/NA nm$

BTA-1 is an uncharged derivative of thioflavin-T that has high affinity for Ab fibrils and shows very good brain entry and clearance. The Kd of [3H]BTA-1 for binding to AD brain is very similar to the Kd for binding to synthetic Ab fibrils. BTA-1 does not appear to bind significantly to common neuroreceptors or transporter sites. BTA-1 exhibits high affinity for amyloid deposits (Ki = 11 nM for Ab(1-40)). It crosses the blood brain barrier and displays up to 50-fold higher affinity than ThT. It selectively stains cerebral plaques and cerebrovascular amyloid deposits in the brains of PS1/APP transgenic mice.

Reference:

Ishikawa K, et al. (2004). Amyloid imaging probes are useful for detection of prion plaques and treatment of transmissible spongiform encephalopathies. J Gen Virol 85, 1785-90. Klunk WE, et al. (2003).

Name :	BTA-2 [2-(4"-(dimethylamino)phenyl)-6-methyl-benzothiazole] [L]
Catalog Number :	FP-BS6490 100 mg
	MW: 268.4
	$\lambda_{\rm exc} \lambda_{\rm em} = 356/437 \ \rm nm$

BTA-2 is an uncharged derivative of thioflavin-T that exhibits high affinity for amyloid deposits (Ki = 143 nM for A??(1-40)) and can cross the blood brain barrier. It displays up to 6-fold higher affinity than ThT and stains both plaques and neurofibrillary tangles in post mortem Alzheimer disease brain.

References:

Mathis CA, et al. (2002). A lipophilic thioflavin-T derivative for positron emission tomography (PET) imaging of amyloid in brain. Bioorg Med Chem Lett 12, 295-8. Kung HF, et al. (2001). Novel stilbenes as probes for amyloid plaques. J Am Chem Soc 123, 12740-1.

Name :	Chrysamine G
Catalog Number :	FP-BS6500 10 mg
	MW:482.5; Soluble in DMSO
	$\lambda_{exc} \lambda_{em} = 386/NA nm$

Chrysamine G (CG) is a carboxylic acid analog of Congo red, a histologic dye which stains amyloid. CG binds to the bamyloid protein of Alzheimer's disease (AD) in vitro and partitions into the brain of normal mice. The binding of CG is correlated with numbers of senile plaques and neurofibrillary tangles. CG displays both high (Kd = 200 nM; Bmax = 1.13 moles per mole of Ab40) and low (Kd = 38.77 mM; Bmax = 23.10 moles per mole of Ab40) affinity binding sites for bamyloid (Ab) fibrils. It can cross the blood-brain barrier and serve as an useful probe for detecting senile plaques (Ab aggregate). In addition, CG can be used to stain cerebrovascular amyloid in tissue sections.

References:

Mathis CA, et al. (2004). Imaging b-amyloid plaques and neurofibrillary tangles in the aging human brain. Curr Pharm Des 10, 1469-92. Klunk WE, et al. (1998). Chrysamine-G, a lipophilic analogue of Congo red, inhibits A?-induced toxicity in PC12 cells. Life Sci 63, 1807-14. Klunk WE, et al. (1995). Chrysamine-G binding to Alzheimer and control brain: autopsy study of a new amyloid probe. Neurobiol Aging 16, 541-8.

Name :	Half Chrysamine G (hCG)
Catalog Number :	FP-BS6520, 10 mg
	MW = 242.2
	Soluble in DMSO
	$\lambda_{exc} = 342/NA nm$

The neurotoxicity of Ab is widely believed to play a seminal role in neurodegeneration in Alzheimer's disease. Half chrysamine G (hCG) has a lower affinity for Ab compared with that of CG. Both CG and hCG are equally efficacious in reducing Ab-induced neuronal death at a concentration of 0.1-1 mM, indicating that the mechanism of action for CG is not due to its chelating activity, but rather due to its anti-oxidant activity.

Reference:

Ishii K, et al. (2002). Chrysamine G and its derivative reduce amyloid ?-induced neurotoxicity in mice. Neurosci Lett 333, 5-8.



FT-CG2370

Name :	Congo Red (CR)
Catalog Number :	"UltraPure Grade" N12511 1 g "Solution" FP-AQ3370 100 tests MW : 696.67 ; Soluble in water $\lambda_{exc} \lambda_{em} = 610/N/A, C_{24}H_{17}FO_6$

Early diagnosis and classification of amyloid deposition and differentiation from other glomerular fibrillar deposits rely on routine **Congo red** (CR) histochemistry. CR binding, monitored by characteristic yellow-green birefringence under crossed polarization has been used as a diagnostic test for the presence of amyloid in tissue sections for several decades. This assay is also widely used for the characterization of in vitro amyloid fibrils.

CR is sandwiched between two protein molecules causing protein oligomerization. Congo red fluorescence (CRF) is an alternative method based on examination of the CR-stained section by ultraviolet (UV) light. CRF is simple to perform and more pronounced, therefore easier to evaluate than CR in bright light. Congo red, when combined with immunohistochemistry, is still visible under UV whereas CR is masked in bright light.

Although not widely used, the CRF method for detecting amyloid is simple to use with a high specificity and sensitivity, and may be applied successfully to frozen sections.

References:

Tzankov A, et al. (2003). Congo red-positive cardiac kappa-AL amyloidosis in plasmacytoma ?Xcase report and review of the literature. Acta Med Austriaca 30, 29-32 ; Roterman I, et al. (2001). Why Congo red binding is specific for amyloid proteins - model studies and a computer analysis approach. Med Sci Monit 7, 771-84 ; Khurana R, et al. (2001). Is Congo red an amyloid-specific dye? J Biol Chem 276, 22715-21.

Name :	ThioFlavin T
Catalog Number :	FP-BS6530, 1g
	MW: 318.9
	Soluble in DMSO
	$\lambda_{exc} = 412/482nm$

The benzothiazole dye **thioflavin T** (ThT) is a classic amyloid stain for senile plaques containing bA4 peptide in Alzheimer's disease brain. ThT also binds rapidly and specifically to the anti-parallel b-sheet fibrils formed from synthetic b-amyloid (1-40), but does not bind to monomer or oligomeric intermediates. The fibrillar b-sheet-bound dye species undergoes a characteristic 120 nm red shift of its excitation spectrum that may be selectively excited at 450 nm, resulting in a fluorescence signal at 482 nm. ThT is a useful probe for the aggregated fibrillar state of b-amyloid (1-40) fibrils as the amyloid-specific fluorescence reports only fibrillar species. The binding of ThT does not interfere with the aggregation of this peptide into amyloid fibrils. The putative conformational changes detected by the ThT fluorescence suggest that small pharmacologic ligands can perturb and possibly dissociate Ab amyloid fibrils.

References:

Ban T, et al. (2003). Direct observation of amyloid fibril growth monitored by thioflavin T fluorescence. J Biol Chem 278, 16462-5. Kung MP, et al. (2002). IMPY: an improved thioflavin-T derivative for in vivo labeling of ß-amyloid plaques. Brain Res 956, 202-10. De Ferrari GV, et al. (2001). Thioflavin T is a fluorescent probe of the acetylcholinesterase peripheral site that reveals conformational interactions between the peripheral and acylation sites. J Biol Chem 276, 23282-7.

Ordering information

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

For any information, please ask : FluoProbes® / Interchim; Hotline : +33(0)4 70 03 73 06

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