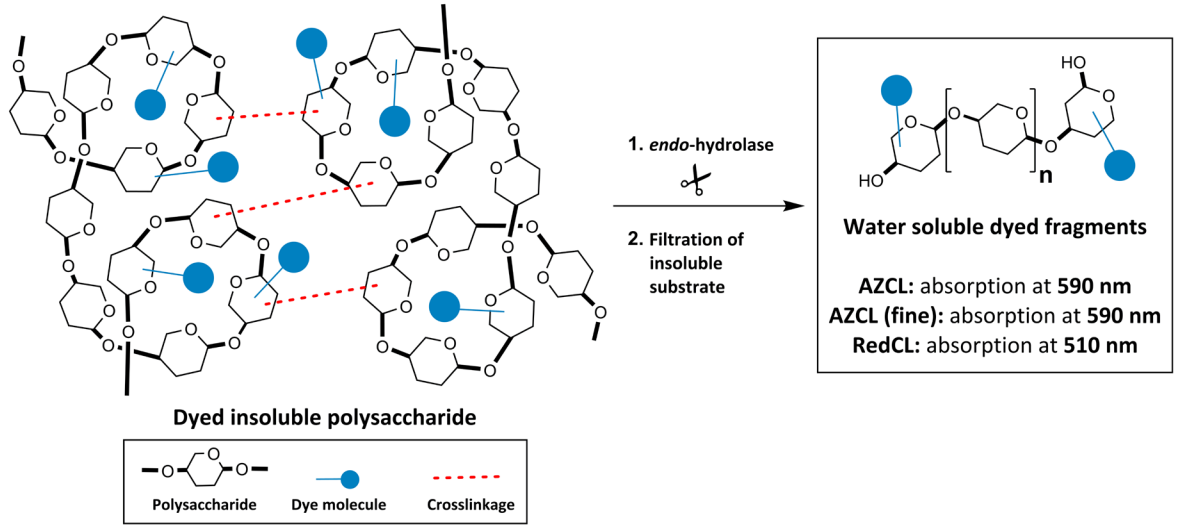
INSOLUBLE DYED POLYSACCHARIDES FOR THE ASSAY OF *endo*-HYDROLASES

06/21

Insoluble dyed polysaccharides are prepared by dyeing and crosslinking highly purified polysaccharides. These substrates are insoluble in buffered solutions, but rapidly hydrate to form gel particles which are readily and rapidly hydrolysed by specific *endo*-hydrolases, releasing soluble dye-labelled fragments.



These substrates can be used to detect enzymatic activities in agar plate, microtiter plate assays and semi-quantitative test tube assays. These methods allow for high throughput screening of multiple samples and are accurate, cost effective and easily performed.

Megazyme supplies three types of insoluble dyed polysaccharides:

Type	Form	Particle size	Absorbance	Colour
AZCL-polysaccharides	Granular	< 0.5 mm	590 nm	Blue/black
AZCL-polysaccharides (fine)	Fine powder	~ 0.1 mm	590 nm	Blue/black
RedCL-polysaccharides (fine)	Fine powder	~ 0.1 mm	510 nm	Red

The traditionally supplied substrates (AZCL-polysaccharides) are also available in tablet form. They can be used to measure enzyme activities in standard test-tube assays.

Recommended methods for screening enzymatic activities using insoluble dyed substrates are detailed in the Application Note, which is available under the Documentation tab.

SUBSTRATES, APPLICATIONS and SUGGESTED BUFFER

SUBSTRATE	CAT. NO.	ENZYME	BUFFER
AZCL-Amylose	I-AZAMY	α-Amylase (Fungal)	Na acetate, 100 mM, pH 4.4.
AZCL-Barley β-Glucan	I-AZBGL	Malt β-Glucanase	Na acetate, 25 mM, pH 4.5.
AZCL-HE-Cellulose	I-AZCEL	*endo*-Cellulase (*Trichoderma*)	Na acetate, 25 mM, pH 4.5.
AZCL-Pachyman AZCL-Curdlan	I-AZPAC I-AZCUR	*endo*-1,3-β-Glucanase	Na acetate, 50 mM, pH 6.0.
AZCL-Arabinan (Debranched)	I-AZDAR	*endo*-1,5-α-Arabinanase	Na acetate, 50 mM, pH 4.0.
AZCL-Dextran	I-AZDEX	*endo*-1,6-α-Dextranase	Na acetate, 50 mM, pH 5.0.
AZCL-Galactomannan	I-AZGMA	*endo*-1,4-β-Mannanase	Na acetate, 50 mM, pH 4.5.
AZCL-Galactan (Potato)	I-AZGLP	*endo*-1,4-β-Galactanase	Na acetate, 25 mM, pH 4.3.
AZCL-Chitosan	I-AZCHAN	Chitosanase	Na acetate, 50 mM, pH 5.0.
AZCL-Pullulan	I-AZPUL	Malt Limit-dextrinase	Na maleate, 100 mM, pH 5.5.
AZCL-Xyloglucan (Tamarind)	I-AZXYG	*endo*-Cellulase (*Trichoderma*)	Na acetate, 25 mM, pH 4.5.
AZCL-Xylan (Beechwood) AZCL-Arabinoxylan (Wheat) AZCL-Arabinoxylan (Wheat) (fine)	I-AZXBE I-AZWAX I-AZWAXF	*endo*-1,4-β-Xylanase	Na acetate, 25 mM, pH 4.7.
AZCL-Casein AZCL-Collagen	I-AZCAS I-AZCOL	*endo*-Protease	Na phosphate, 100 mM, pH 7.0.
AZCL-Rhamnogalacturonan I	I-AZRHI	Rhamnogalacturonan hydrolase and lyase	Na acetate, 50 mM, pH 4.5 (or 8).

CONDITIONS OF USE OF AZCL-POLYSACCHARIDES:

AZCL-Polysaccharides are the active ingredient in the test tablets supplied by Megazyme. Consequently, further information on possible applications and assay conditions for these substrates can be obtained by reference to the appropriate Test Tablet assay protocol. In general, the concentration of the AZCL-polysaccharide in the test tablet is 30% w/w.

AZCL-Polysaccharide	Test Tablet	For the Measurement of:
AZCL-Amylose	Amylazyme	α-Amylase
AZCL-Barley β-Glucan	β-Glucazyme	Malt β-Glucanase Lichenase *endo*-Cellulase (*Trichoderma*)
AZCL-HE-Cellulose	Cellazyme C	*endo*-Cellulase (*Trichoderma*)
AZCL-Chitosan	Chitozyme	*endo*-Chitosanase
AZCL-Curdlan	1,3-β-Glucazyme HS	*endo*-1,3-β-Glucanase
AZCL-Arabinan (Debranched)	Arabinazyme	*endo*-1,5-α-Arabinanase
AZCL-Dextran	α-Dextrazyme	*endo*-1,6-α-Dextranase
AZCL-Galactomannan	Mannazyme	*endo*-1,4-β-Mannanase
AZCL-Galactan (Potato)	Galactazyme	*endo*-1,4-β-Galactanase
AZCL-Pullulan	Limit-Dextrizyme	Malt Limit-dextrinase Microbial pullulanase
AZCL-Xyloglucan (Tamarind)	Cellazyme T	*endo*-Cellulase (*Trichoderma*)
AZCL-Xylan (Birchwood) AZCL-Xylan (Beechwood)	(refer to Xylazyme AX)	*endo*-1,4-β-Xylanase
AZCL-Arabinoxylan (Wheat)	Xylazyme Xylazyme AX	*endo*-1,4-β-Xylanase
AZCL-Casein I-AZCAS	Protazyme AK	*endo*-Protease
AZCL-Collagen I-AZCOL	Protazyme OL	*endo*-Protease