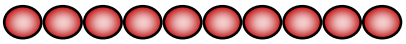
PECTIC POLYSACCHARIDES

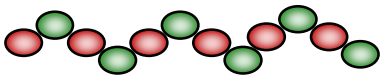
Galacturonic acid - rich polysaccharides from the primary plant cell wall matrix

Galacturonic acid (GalA) occurs in two core domains that may be linked covalently :

1 - α(1→4)-linked D-GalA : **HOMOGALACTURONAN (HG)**

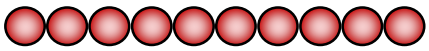


2 - a repeating dimer of →4)GalAα(1→2)Rhaα(1→ : **RHAMNOGALACTURONAN (RG)**



HG & RG are subject to modifications/substitutions → altered properties & functions

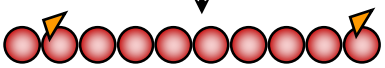
HOMOGALACTURONAN



synthesized in methyl-esterified form

de-methylesterification *in muro* by pectin methyl esterases

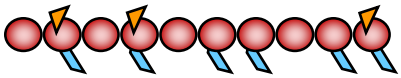
blockwise



non-blockwise

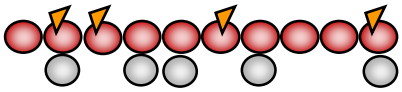
HG

acetylation



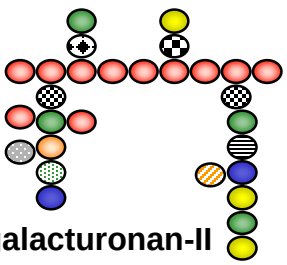
acetylated HG

xylose substitution



xylogalacturonan (XG)

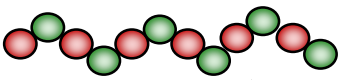
complex side chains



rhamnogalacturonan-II (RG-II)

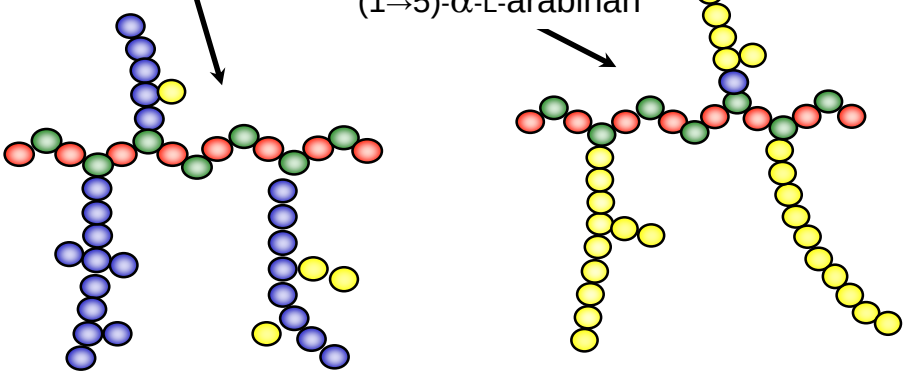
methyl acetyl

RHAMNOGALACTURONAN (RG)



(1→4)-β-D-galactan

(1→5)-α-L-arabinan



rhamnogalacturonan-I (RG-I)

20-80% of Rha residues in RG are substituted with neutral side chains rich in Gal and Ara. The GalA in the backbone can be methyl-esterified, acetylated etc. as in HG

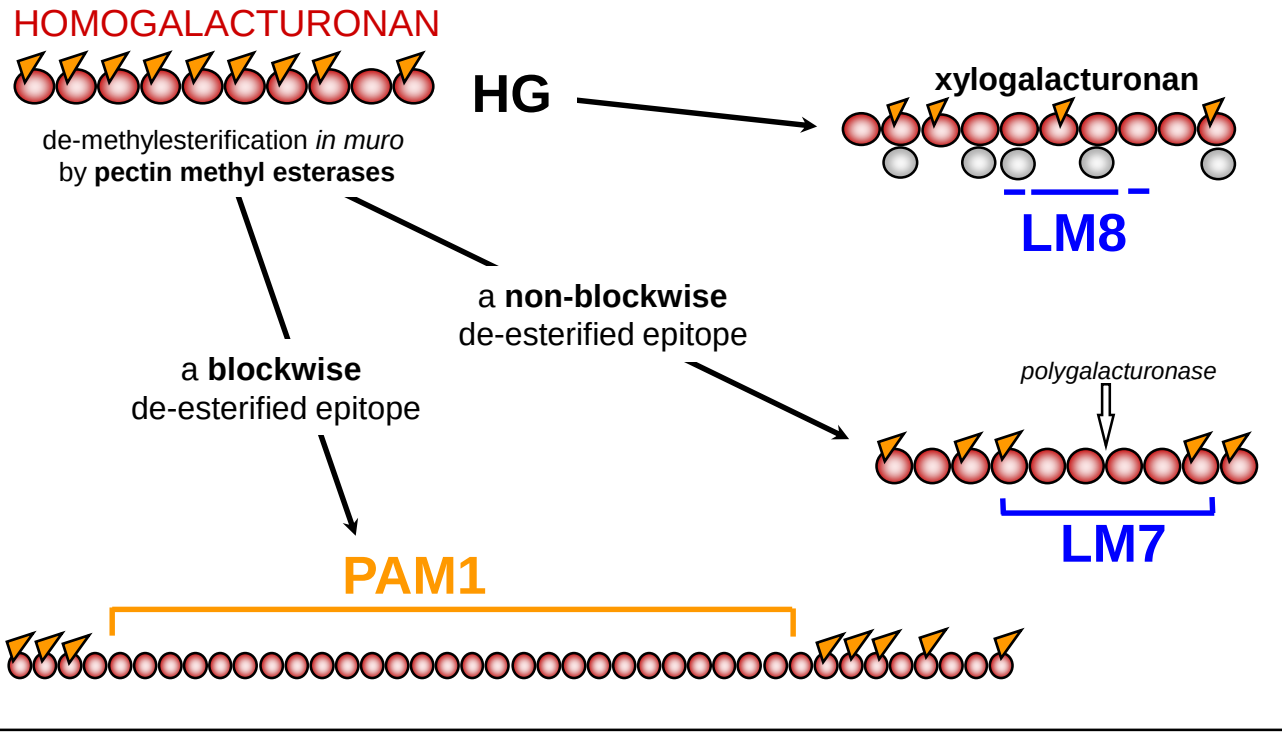
GalA
Rha
Ara
Gal
Xyl
Api
KDO
Dha
GlcA
Fuc
AcMeFuc
MeXyl
AcAce

Paul Knox 11/2019

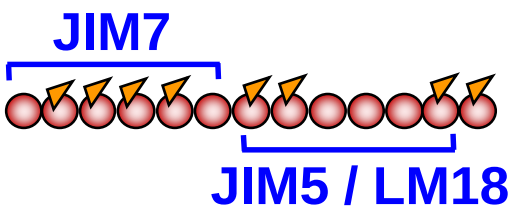
MONOCLONAL ANTIBODIES TO PECTIC POLYSACCHARIDES

Pectic polysaccharides are structurally complex and antibodies are useful to determine the distribution and organization of pectic domains within the primary cell wall matrix & their dynamics during plant processes.

Our antibodies are mostly made using **hybridoma** technologies. A range of monoclonal antibodies to pectic polysaccharide epitopes have been generated.

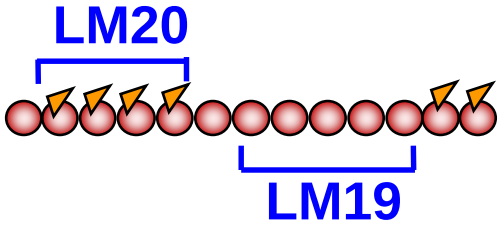


The once widely used **JIM5** and **JIM7 HG probes** recognise a range of partially methyl-esterified HG structures with representative epitopes shown below.



More recent MAbs **LM19** and **LM20** have clearer discrimination of methylesterified and un-esterified HG with representative epitopes shown below. LM20 recognizes HG with a high density of methylesters.

LM19 is our best MAb for unesterified HG.

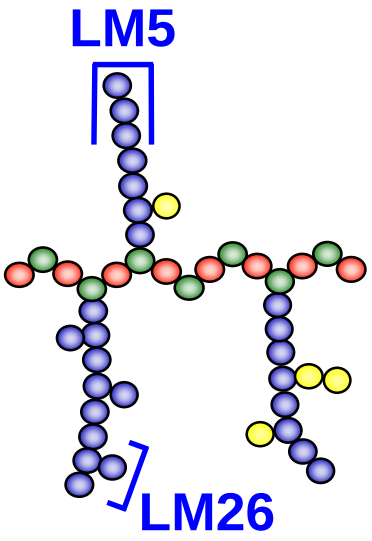


For the MAb-based analysis of pectic HG we now recommend the combined use of **LM19** and **LM20** along with **JIM7** – the latter having a wide recognition of methylester epitopes and strong recognition of HG in general. **LM18** has a similar recognition profile to **JIM5**.

RHAMNOGALACTURONAN-I (RG-I)

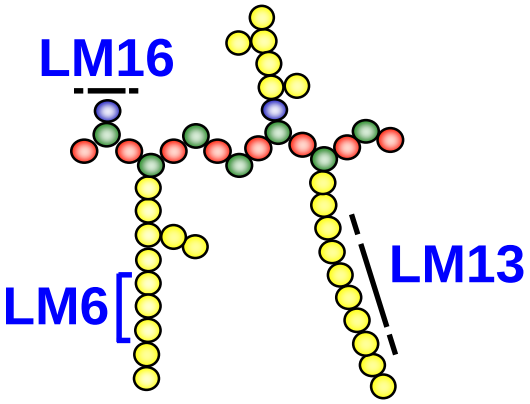
Antibodies to epitopes occurring in the neutral side chains of RG-I have been prepared using neoglycoproteins & also isolated subsequent to complex immunogens.

(1→4)-β-D-galactan



It is now known that **LM5** binds to the non-reducing end of pectic galactan. The recently isolated MAb **LM26** binds to a branched epitope of pectic galactan with a 1,6-galactosyl residue on the 1,4-galactan backbone.

(1→5)-α-L-arabinan



LM6 (and also **LM6-M** (an IgM)) bind to several residues of 1, 5-arabinan and **LM13** binds to longer linear regions of arabinan. The proposed epitope of **LM16** is a galactosyl stub on RG backbone that is created by arabinan removal.

Epitope defined

Epitope not yet defined

M Hybridoma monoclonal antibody

M Phage display monoclonal antibody

Major pectic MAb References

Homogalacturonan

Clausen *et al*. (2004) **Carbohydrate Research** 338, 1797-1800 ONLINE
Verhertbruggen *et al*. (2009) **Carbohydrate Research** 344, 1858-1862 ONLINE
Marcus *et al*. (2010) **Plant Journal** 64, 191-203 ONLINE ***– see Fig. 5***

Rhamnogalacturonan-I side chains

Galactan

Andersen *et al*. (2016) **Carbohydrate Research** 436, 36-40 ONLINE
Torode *et al*. (2018) **Plant Physiology** 176, 1547-1558 ONLINE

Arabinan

Verherbruggen *et al*. (2009) **Plant Journal** 59, 413-425 ONLINE
Cornuault *et al*. (2017) **BioRχiv** ONLINE

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