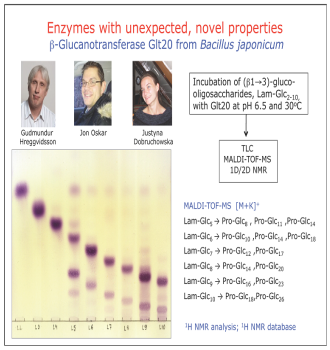


Enzymes with Novel Properties

Description

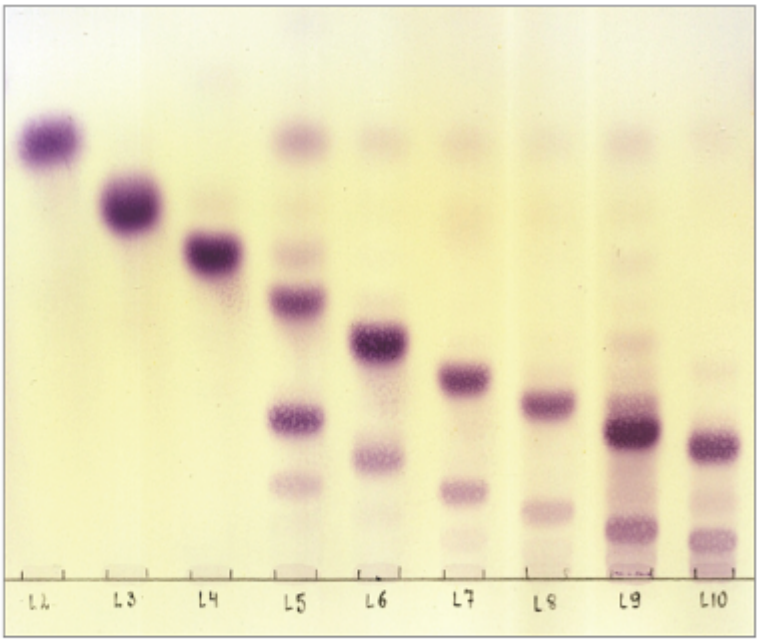
Enzymes with unexpected, novel propert

β-Glucanotransferase Glt20 from *Bacillus japoni*

Gudmundur Hreggvidsson

Jon Oskar

Justyna Dobruchowska



Incubation of (β1→3
oligosaccharides, La
with Glt20 at pH 6.5

↓

TLC
MALDI-TOF-M
1D/2D NMR

MALDI-TOF-MS [M+K]⁺

Lam-Glc_5 → Pro-Glc_8 , Pro

Lam-Glc_6 → Pro-Glc_{10} ,Pro

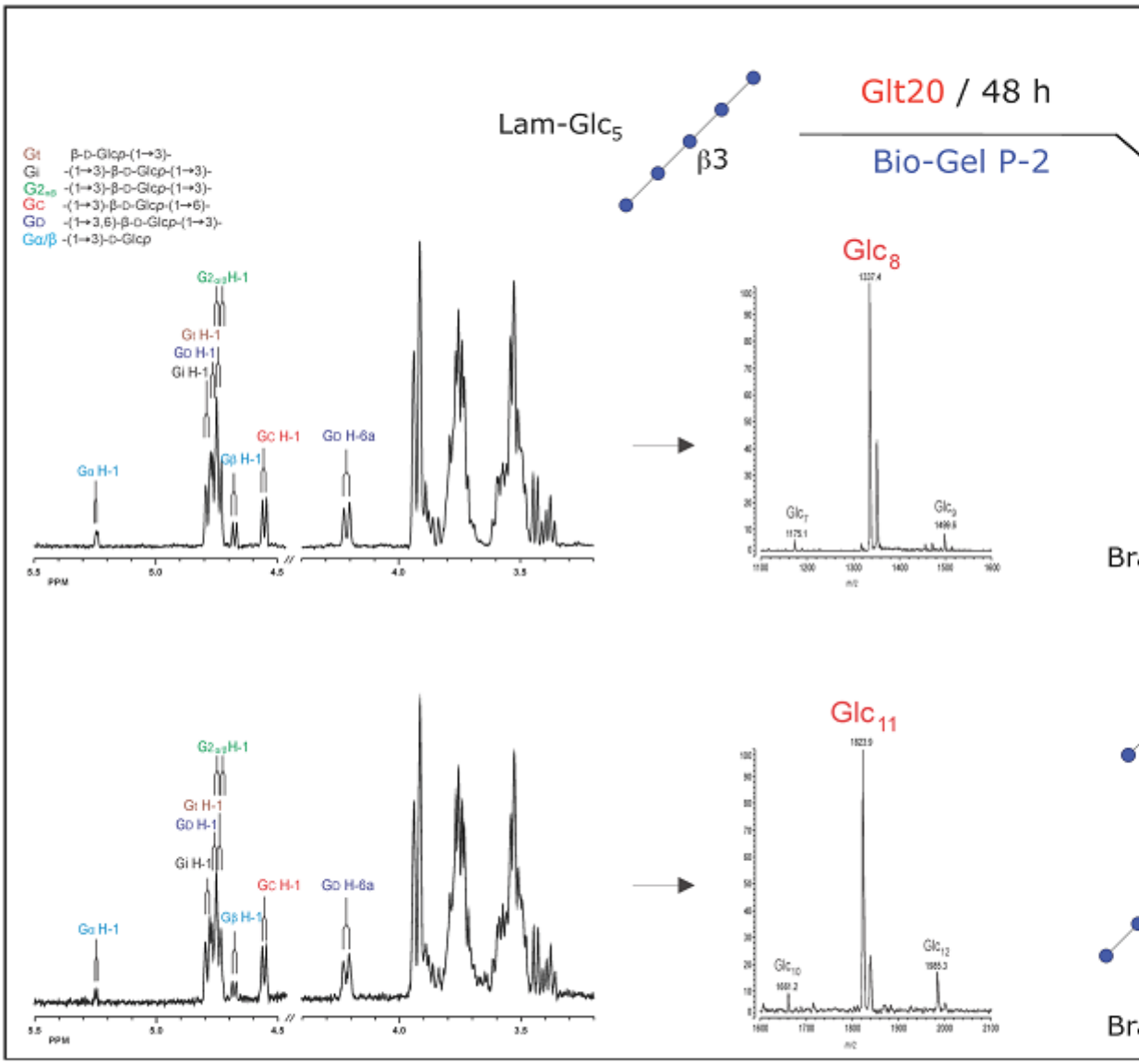
Lam-Glc_7 → Pro-Glc_{12} ,Pro

Lam-Glc_8 → Pro-Glc_{14} ,Pro

Lam-Glc_9 → Pro-Glc_{16} ,Pro

Lam-Glc_{10} → Pro-Glc_{18},Pro

^{1}H NMR analysis; ^{1}H NMR


Lam-Glc5
β3
Glt20 / 48 h
Bio-Gel P-2
Gt β-D-Glcp-(1→3)-
Gi -(1→3)-β-D-Glcp-(1→3)-
G2n5 -(1→3)-β-D-Glcp-(1→3)-
Gc -(1→3)-β-D-Glcp-(1→6)-
GD -(1→3,6)-β-D-Glcp-(1→3)-
Gα/β -(1→3)-D-Glcp
G2α/βH-1
Gt H-1
GD H-1
Gi H-1
Gc H-1
GD H-6a
Gβ H-1
Gα H-1
PPM
Glc8
Glc7
Glc9
Glc11
Glc10
Glc12
Br
Br

Bio-active (1→3,1→6)-β-D-glucans

The immunostimulating properties of (β1→3)-gluca
with varying numbers of (β1→6) branches have bee
recognized by decades.

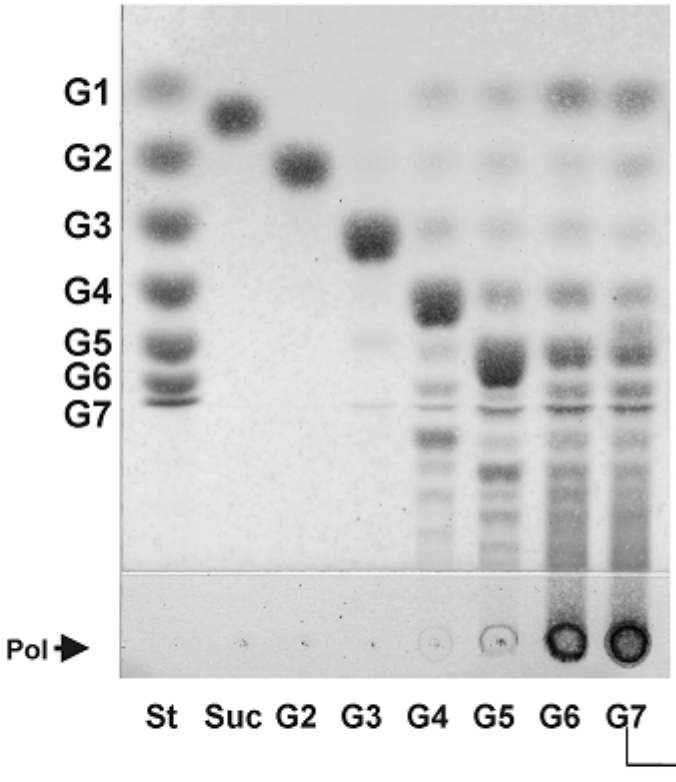
The wide of affinities appears to be due to the diffe
sizes and numbers of branches in the (β1→3,β1→6)
glucans.

Using Glt20, linear (β1→3)-glucans can be converte
mixtures of (β1→3,β1→6)-glucans, with major amo
of multiple branched structures.

Immunological evaluations of the product(s)(mixtu
are underway.

Enzymes with unexpected, novel propert

4,6-α-Glucanotransferase GTFB of *Lactobacillus reuteri*



Incubation of 90
with 25 mM sucr
mM malto-oligosa

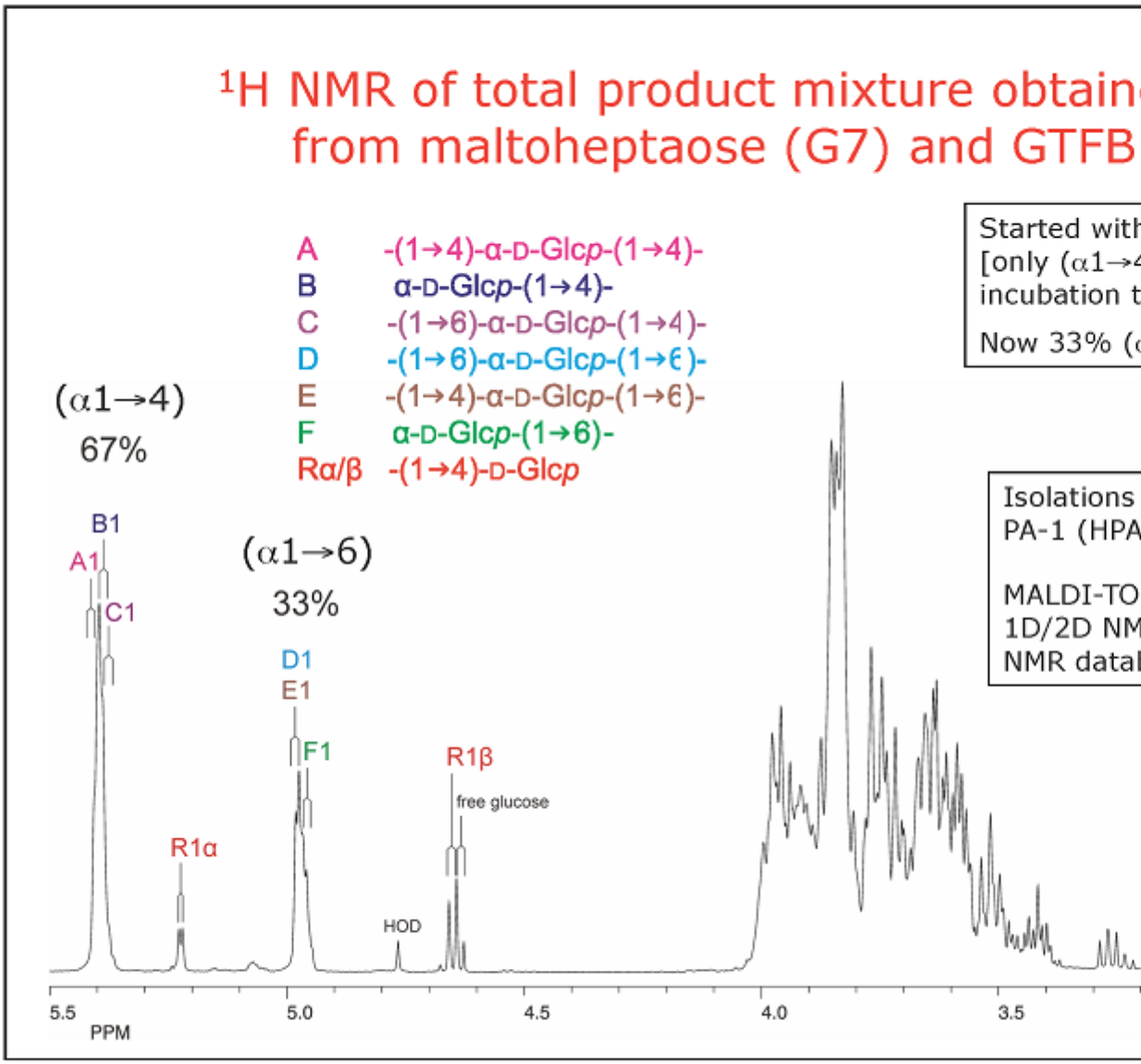
TLC analysis of th
products.

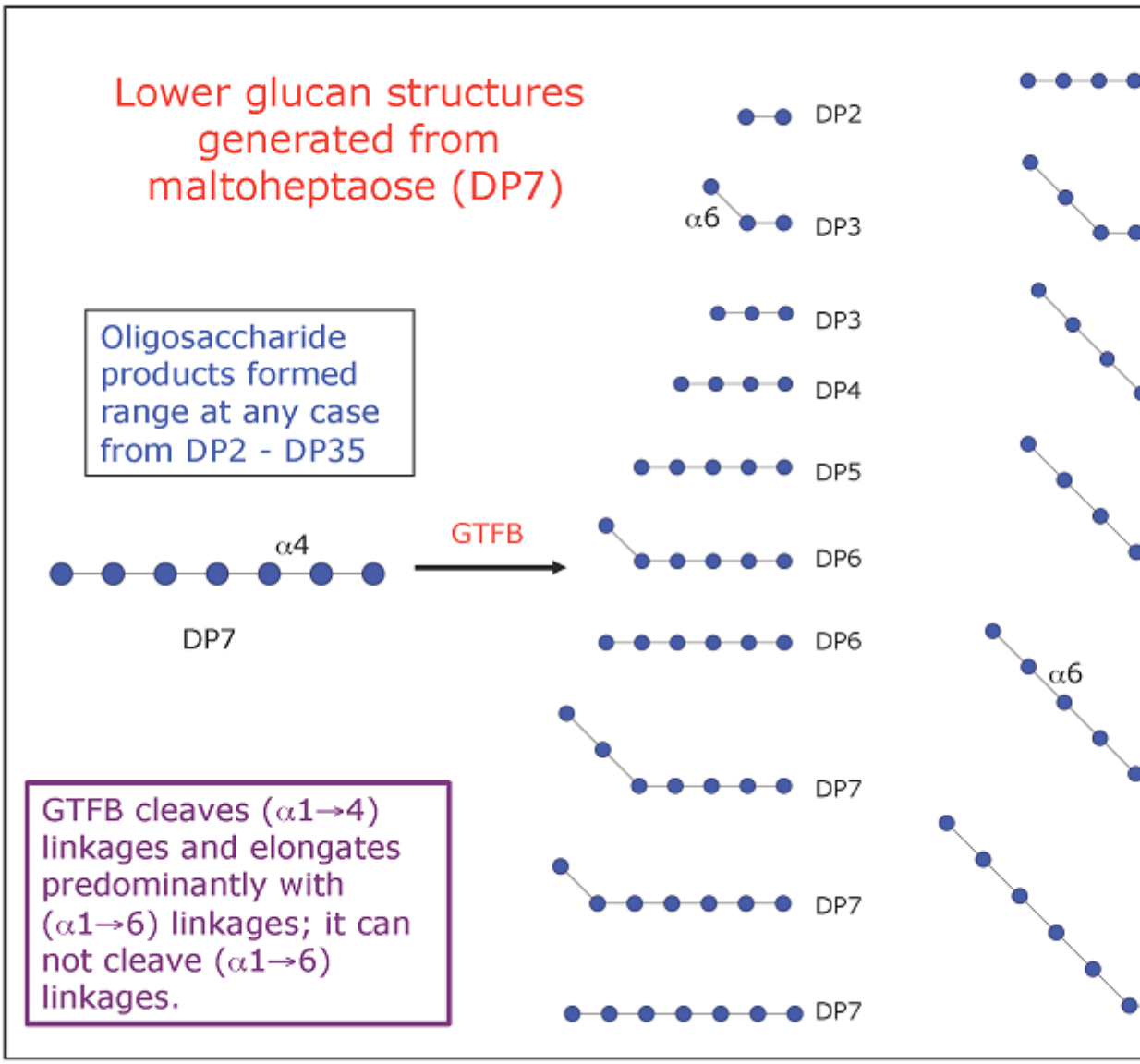
Lubbert Dijkhuizen

^{1}H NMR analysis

Reaction conditions: 13 h, 37°C / 50 mM NaOAc buffer, pH 4.7 / 1 mM $CaCl_2$

St = standards; Suc = sucrose; G1 = glucose; G2 = maltose; G3 = r
G4 = maltotetraose; etc.


¹H NMR of total product mixture obtain
from maltoheptaose (G7) and GTFB
Started with
[only (α1→4
incubation t
Now 33% (α
A -(1→4)-α-D-Glcp-(1→4)-
B α-D-Glcp-(1→4)-
C -(1→6)-α-D-Glcp-(1→4)-
D -(1→6)-α-D-Glcp-(1→6)-
E -(1→4)-α-D-Glcp-(1→6)-
F α-D-Glcp-(1→6)-
Rα/β -(1→4)-D-Glcp
(α1→4)
67%
(α1→6)
33%
B1
A1
C1
D1
E1
F1
R1α
R1β
free glucose
HOD
Isolations
PA-1 (HPA
MALDI-TO
1D/2D NM
NMR datal
5.5
PPM
5.0
4.5
4.0
3.5


Lower glucan structures generated from maltoheptaose (DP7)
Oligosaccharide products formed range at any case from DP2 - DP35
α4
DP7
GTFB
DP2
α6
DP3
DP3
DP4
DP5
DP6
DP6
α6
DP7
DP7
DP7
GTFB cleaves (α1→4) linkages and elongates predominantly with (α1→6) linkages; it can not cleave (α1→6) linkages.

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Glycobiology vol. 22 no. 4 pp. 517–528, 2012
doi:10.1093/glycob/cwr167
Advance Access publication on December 2, 2011

Structural characterization of linear isomalto-/malto oligomer products synthesized by the novel GTFB 4 α-glucanotransferase enzyme from *Lactobacillus reuteri* 121

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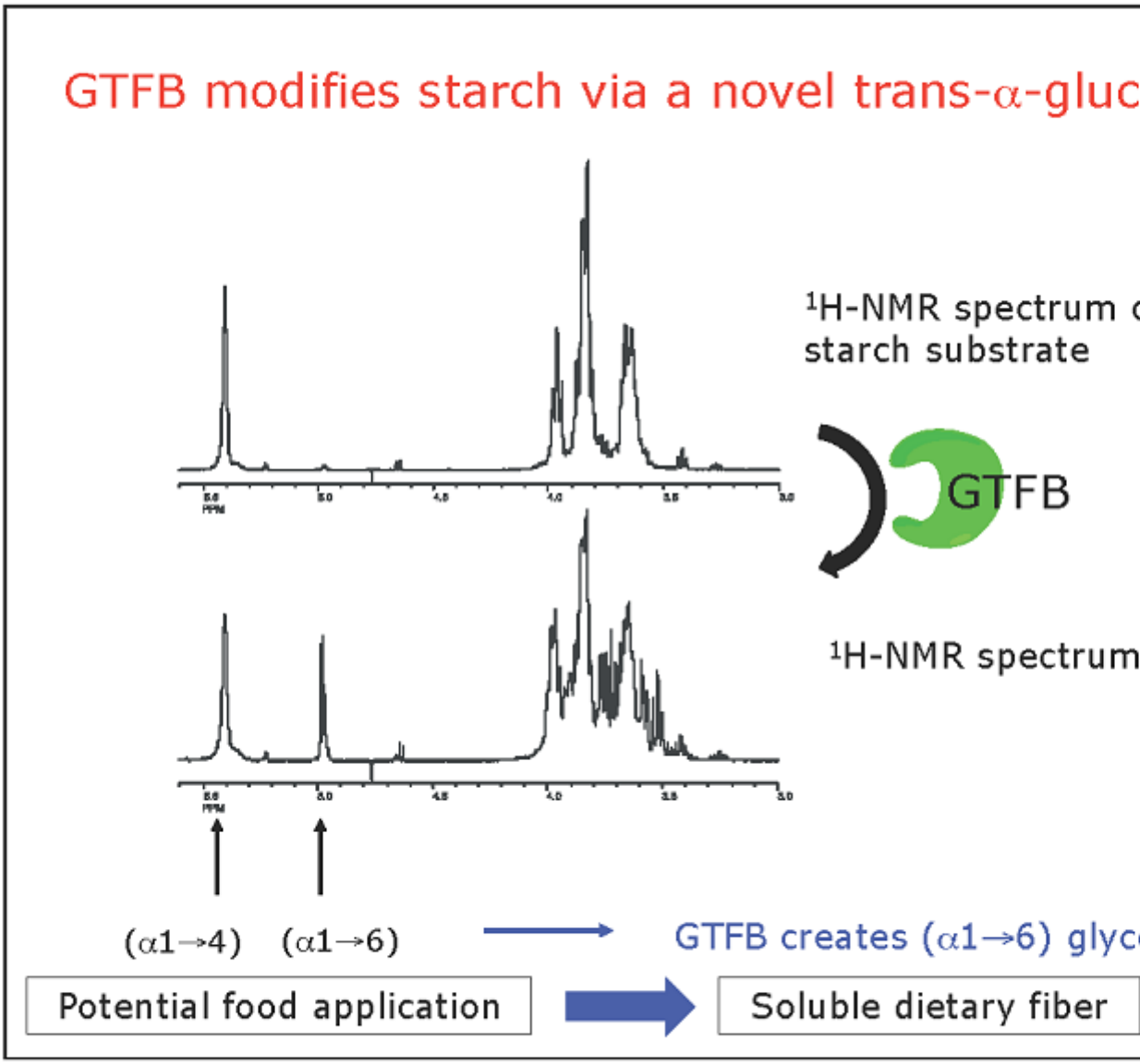
Received on September 28, 2011; revised on November 14, 2011; accepted on November 15, 2011

emulsifying, sweetening, gelling or wate
well as their prebiotic properties. LAB
glucansucrases (GSs)/glucosyltransferases
to convert their natural substrate sucrose
EPSs, being complex α-D-glucose poly
strains possess multiple GTF enzymes.

When searching for novel carbohydrate-
which may be used in industrial applicatio
several *gtf* genes (e.g. *gtf4*, *gtf180*, *gtf*



Category

1. News