

Supporting Information

Specific interactions of leucine with disaccharides by electrospray ionization mass spectrometry: Application for rapid differentiation of disaccharide isomers in combination with statistical analysis

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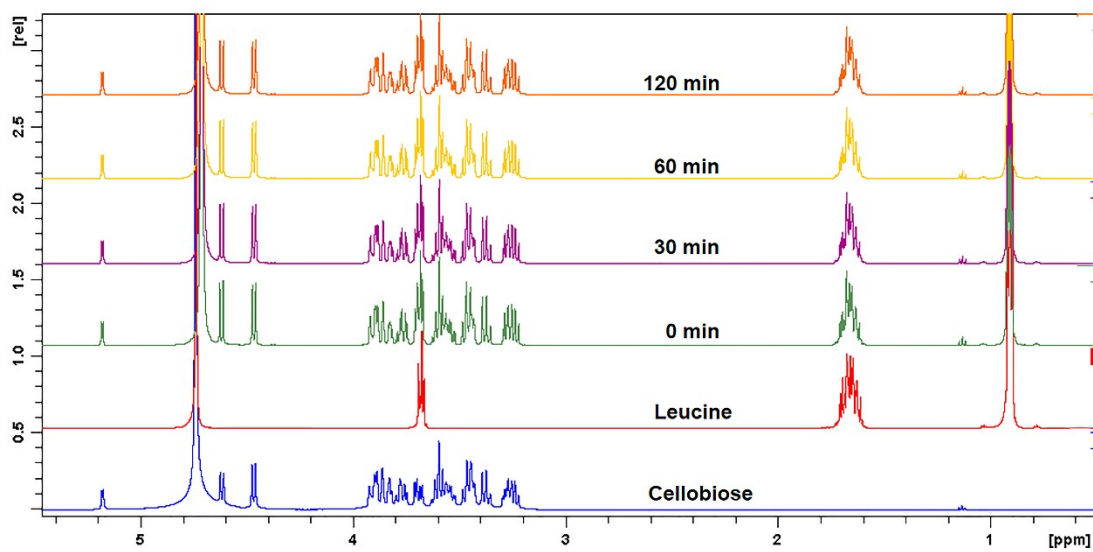


Fig. S1 The stacked ¹H NMR spectra for cellobiose (1 mmol) incubated with leucine (1 mmol) and KCl (0.1 mmol) in D₂O at room temperature.

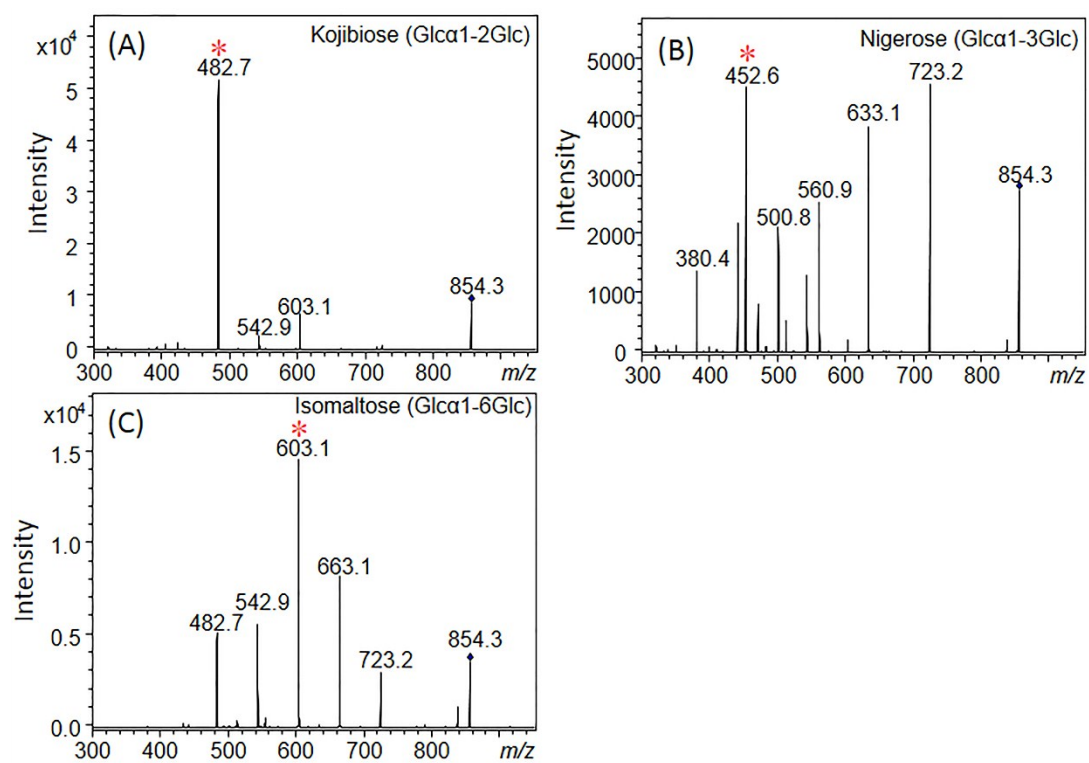


Fig. S2 ESI-IT-MS/MS spectra of complexes $[2\text{Dis}+\text{Leu}+\text{K}]^+$ for three glycopyranosyl-glucose disaccharides in positive ion mode. (A) Kojibiose ($\text{Glc}\alpha 1\text{-}2\text{Glc}$); (B) Nigerose ($\text{Glc}\alpha 1\text{-}3\text{Glc}$); (C) Isomaltose ($\text{Glc}\alpha 1\text{-}6\text{Glc}$). The typical fragment ions were marked by “*”.

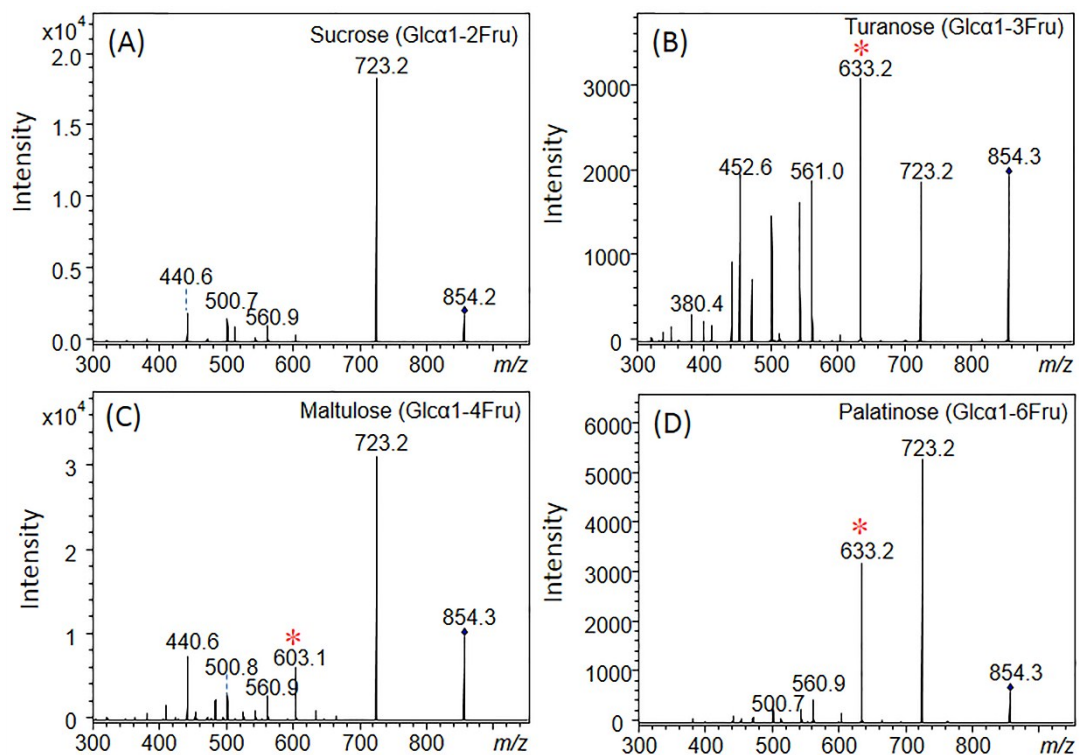


Fig. S3 ESI-IT-MS/MS spectra of [2Dis+Leu+K]⁺ complexes for four glucopyranosyl-fructose disaccharides in positive ion mode. (A) Sucrose (Glc α 1-2Fru); (B) Turanose (Glc α 1-3Fru); (C) Maltulose (Glc α 1-4Fru); (D) Palatinose (Glc α 1-6Fru). The typical fragment ions were marked by “*”.

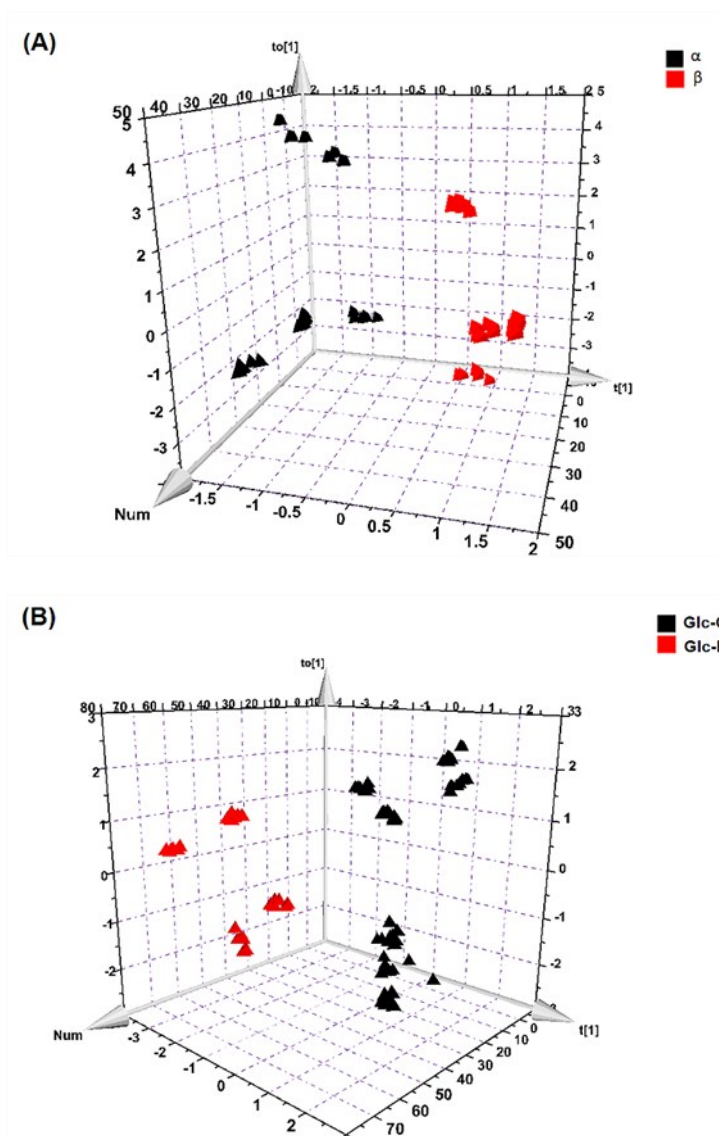


Fig S4 OPLS-DA analysis of the complexes $[2\text{Dis}+\text{Leu}+\text{K}]^+$ comprised by twelve glucose-containing disaccharides by Q-TOF-MS/MS. (A) Discrimination of α isomers from β isomers; (B) Discrimination of Glc-Glc from Glc-Fru with different mono units.

Table S1 Formation of ternary Cello/AA/K⁺ complexes using ESI-IT-MS and Q-TOF-MS respectively.

AA ^a	Cello+2AA+K ⁺	Relative Abundance (%)		2Cello+AA+K ⁺	Relative Abundance (%)	
	Calculated Mass	IT-MS	Q-TOF-MS	Calculated Mass	IT-MS	Q-TOF-MS
Ala	559.2	n.d. ^b	n.d.	812.2	n.d.	n.d.
Arg	729.3	n.d.	n.d.	897.3	n.d.	n.d.
Asn	645.2	n.d.	n.d.	855.2	n.d.	n.d.
Asp	647.2	n.d.	n.d.	856.2	n.d.	n.d.
Cys	623.1	n.d.	n.d.	844.2	n.d.	n.d.
Glu	675.2	n.d.	n.d.	870.2	n.d.	n.d.
Gln	673.2	n.d.	n.d.	869.3	n.d.	n.d.
Gly	531.1	n.d.	n.d.	798.2	n.d.	n.d.
His	691.2	n.d.	n.d.	878.3	n.d.	n.d.
Ile	643.3	10±4	8±3	854.3	n.d.	n.d.
Leu	643.3	15±2	12±3	854.3	25±5	20±6
Lys	673.3	n.d.	n.d.	869.3	n.d.	n.d.
Met	679.2	n.d.	n.d.	872.2	n.d.	n.d.
Phe	711.2	n.d.	n.d.	888.3	n.d.	n.d.
Pro	611.2	n.d.	n.d.	838.3	n.d.	n.d.
Ser	591.2	25±8	n.d.	828.2	n.d.	n.d.
Thr	619.2	n.d.	n.d.	842.3	n.d.	n.d.
Trp	789.3	n.d.	n.d.	927.3	n.d.	n.d.
Tyr	743.2	n.d.	n.d.	904.3	n.d.	n.d.
Val	615.2	26±8	n.d.	840.3	n.d.	n.d.

^a AA: amino acid; ^b n.d.: not detected

Table S2 ESI-Q-TOF-MS/MS data of [2Dis+Leu+K]⁺ at *m/z* 854 as the precursor ion.

Structure	Relative abundance of fragment ions (%; Data was exhibited as means S.E.M.)										
	723	663	633	603	561	543	513	512	483	392	381
Glc α 1-2Glc	17 \pm 1			100 \pm 0		12 \pm 1	62 \pm 2		94 \pm 3	6 \pm 1	60 \pm 2
Glc β 1-2Glc	20 \pm 1			83 \pm 3			45 \pm 3		100 \pm 0	10 \pm 1	74 \pm 2
Glc α 1-3Glc	100 \pm 0		36 \pm 4		6 \pm 2	8 \pm 2		46 \pm 0			12 \pm 2
Glc β 1-3Glc	100 \pm 0					12 \pm 1		96 \pm 3			14 \pm 0
Glc α 1-4Glc	69 \pm 4	27 \pm 3		45 \pm 1			76 \pm 2		7 \pm 1		100 \pm 0
Glc β 1-4Glc	72 \pm 3	26 \pm 5		100 \pm 0				54 \pm 5	7 \pm 1		43 \pm 3
Glc α 1-6Glc	61 \pm 3	68 \pm 4		90 \pm 3			88 \pm 4		10 \pm 2		100 \pm 0
Glc β 1-6Glc	79 \pm 1	35 \pm 4		41 \pm 3				100 \pm 0			40 \pm 3
Glc α 1-2Fru	100 \pm 0							41 \pm 3			42 \pm 3
Glc α 1-3Fru	68 \pm 0		100 \pm 0		10 \pm 1	32 \pm 1	46 \pm 4				43 \pm 1
Glc α 1-4Fru	100 \pm 0						77 \pm 4				48 \pm 3
Glc α 1-6Fru	100 \pm 0		31 \pm 1				31 \pm 1				37 \pm 1