SUPPLEMENTARY DATA

Structural comparison of exopolysaccharides from the two-potential probiotic *Limosilactobacillus fermentum* strains MC1 and D12

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Figure S1: Elution profiles of D12 (a) and MC1 (b) EPSs obtained by middle pressure size exclusion chromatography on a Sephacryl S-300 HR column.



Figure S2: ¹H NMR spectra of D12 (a) and MC1 (b) EPSs isolated from the cell surface and extracted with 2 M NaOH. Arrows indicate resonances present in MC1 sample only. Spectra were recorded in D_2O , at 50 °C and 500 MHz.



Figure S3: ¹H NMR spectra of the three fractions, head, center and tail, obtained after size exclusion chromatography of D12 EPS. Spectra were recorded in D_2O , at 50 °C and 500 MHz.



Figure S4: ¹H NMR spectra of the three fractions, head, center and tail, obtained after size exclusion chromatography of MC1 EPS. Spectra were recorded in D₂O, at 50 °C and 500 MHz.



Figure S5: TOCSY spectrum of D12 EPS recorded in D_2O , at 50 °C and 500 MHz. Cross-peaks for spin systems A and B are indicated (as in Table 4).



Figure S6: TOCSY spectrum of D12 EPS recorded in D2O, at 50 °C and 500 MHz. Cross-peaks for spin systems **C** to **G** are indicated (as in Table 4).



Figure S7: HSQC spectrum of MC1 EPS recorded in D_2O , at 50 °C and 500 MHz. Anomeric region is shown in the inset. Cross-peaks for spin systems A to G are indicated (as in Table 4).

Oligosaccharide subjected to chemical shift simulation:

 α -D-Glc $p(1\rightarrow 2)$ - β -D-Galf- $(1\rightarrow 6)[\alpha$ -D-Glc $p(1\rightarrow 2)$]- β -D-Galf- $(1\rightarrow 6)[\alpha$ -D-Glc $p(1\rightarrow 2)$]- β -D-Galf-OMe

Table S1: ¹³C chemical shifts obtained using the simulation tool for 13C nucleus at the Carbohydrate Structure Database (CSDB), the CSDB structural ranking tool and empirical chemical shift simulation.

Position ^a	Residue	Trust	C1	C2	C3	C4	C5	C6
1	β-D-Gal <i>f</i> -OMe	25%	108.9	87.7	76.6	83.5	70.5	70.3
2	α-D-Glcp	93%	99.2	72.5	73.9	70.7	73.5	61.8
3	β-D-Gal <i>f</i>	79%	107.4	87.8	76.6	83.5	70.7	70.4
4	α-D-Glcp	93%	99.2	72.5	73.9	70.7	73.5	61.8
5	β-D-Gal <i>f</i>	69%	107.4	87.8	76.4	83.4	71.3	63.4
6	α-D-Glcp	93%	99.2	72.5	73.9	70.7	73.5	61.8

^a Residue position in the oligosaccharide chain starting from β -D-Galf-OMe

Oligosaccharide subjected to chemical shift simulation:

 β -D-Galf-(1 \rightarrow 6)- β -D-Galf-(1 \rightarrow 6)- β -D-Galf-OMe

Table S2: ¹³C chemical shifts obtained using the simulation tool for 13C nucleus at the Carbohydrate Structure Database (CSDB), the CSDB structural ranking tool and empirical chemical shift simulation.

Position ^a	Residue	Trust	C1	C2	C3	C4	C5	C6
1	β-D-Gal <i>f</i> -OMe	46%	111.4	81.9	77.8	83.7	70.6	70.1
2	β-D-Gal <i>f</i> -OMe	83%	109.4	81.5	78.2	84.6	71.0	70.2
3	β-D-Gal <i>f</i> -OMe	87%	109.6	81.4	78.1	84.3	71.4	63.4

^a Residue position in the oligosaccharide chain starting from β -D-Galf-OMe

Oligosaccharide subjected to chemical shift simulation (3 repeating units):

 $\begin{array}{l} \beta\text{-D-Glc}p\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow3)\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Glc}p(1\rightarrow2)]\text{-}\beta\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}(1\rightarrow6)[\alpha\text{-}D\text{-}Galf\text{-}($

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Position"	Residue	Trust	CI	C2	C3	C4	CS	C6
1	β-D-Gal <i>f</i> -OMe	25%	108.9	87.7	76.6	83.5	70.5	70.3
2	α-D-Glcp	93%	99.2	72.5	73.9	70.7	73.5	61.8
3	β-D-Galf	61%	108.5	80.1	84.9	82.5	70.8	63.3
4	β-D-Glcp	78%	102.9	74.2	83.3	69.3	76.9	61.9
5	β-D-Galf	68%	107.0	87.3	75.8	83.0	69.9	69.7
6	α-D-Glcp	93%	99.2	72.5	73.9	70.7	73.5	61.8
7	β-D-Galf	61%	108.5	80.1	84.9	82.5	70.8	63.3
8	β-D-Glcp	78%	102.9	74.2	83.3	69.3	76.9	61.9
9	β-D-Gal <i>f</i>	68%	107.0	87.3	75.8	83.0	69.9	69.7
10	α-D-Glcp	93%	99.2	72.5	73.9	70.7	73.5	61.8
11	β-D-Gal <i>f</i>	61%	108.5	80.1	84.9	82.5	70.8	63.3
12	β-D-Glep	92%	103.1	73.9	76.4	70.4	76.8	61.6

^a Residue position in the oligosaccharide chain starting from β -D-Galf-OMe