Supplemental Material

Table S1. Primers used to screen for AX(OS)-degrading enzymes in the bifidobacterial strains tested.

Reference sequence	Additional entries used to make consensus	Forward primer (5' → 3')	Reverse primer (5' → 3')	Amplicon length (bp)
BL1543	AAN25334, BAJ67386, EEI80964, BAJ71924, EEQ55939, ADQ02610.	GACAATTCGTTCGCTCAAGT	AGCATGTCATCGGACAGTTT	434
BL1544	AAN25335, BAJ71923, BAJ67385, ACD99106, ADQ02611, EEI80965.	TCTCTTTCTGGAACCACGAG	CATTGGGTTGGTATTCTTCG	433
BL0682	AAN24503, EEI80353, BAJ67558, EEQ54758.	CTACGCCATGACTTCTGACC	AAAGCCTGATTGTGAGCTTG	402
BL0181	AAN24035, EEQ55862, EFV37311, EEI80492, EEG70006, BAF40305, EDN82302, ADH01333, EEP21637, EEB22114, EFM40505, EDT45169.	CACTGGGAYCGCACCATY	CCAGCTGGABGACTGRTAGG	537
BL0544+BL1611	AAN24368, EFV36777, BAJ70234, BAJ71854, AAN25400, EDT88515, EEI80936.	GCAACGAGYTGATGGARG	TCCATATCSACGCCSGAG	451
BL1138	AAN24945, EEI81164, BAJ70635, BAJ66143, ADQ01834, ACJ53056, EEQ54180, ADH00027, EFV36420.	ATCTGGAAAACCTCGGCTAT	ATCCATGAACTGCACCAGAT	437
BL1166	AAN24971, ADG99997, BAJ70606, EEQ54210, ADQ01881, EEI81196.	TATCAAGATGTGGTGCATCG	CGTTCCACTTGTCCGAATAC	434

Table S2. Primers used to sequence the putative *BL1544* gene in the bifidobacterial strains tested. [a)] Position in the genome of *Bifidobacterium longum* subsp. *longum* NCC2705.

Reference sequence	PCR target [a)]	Forward primer (5' → 3')	Reverse primer (5' → 3')	Amplicon length (bp)
*BL1544*_1	1946863-1947441	CTTGCTTGGCTCTCACTGGT	TAATTGGAGATCCGGGACTG	579
*BL1544*_2	1947157-1947846	AAAATTTCGGCTAGTGCTATCG	AGGAGTCTCACCAAGATCATCAG	690
*BL1544*_3	1947693-1948418	TGATCCGTCATTGTATTTTGATG	GTAAGCGTACCATTCCACTGAG	726
*BL1544*_4	1948242-1948977	CAATTCGGACGATTTTGATAATG	GGAAACCTTGTGCCCATTG	736
*BL1544*_5	1948816-1949557	GGTCTTGCTGATGAGAAGGAATA	CGTCGCTCAAGTCATAGTAATCA	742
*BL1544*_6	1949391-1950087	GTACAGTCTGGACGGTAAGCAAT	ACATCACTGATTCCGCACATT	697

Table S3. PC1, PC2, and PC3 coordinates of bifidobacterial strains in three-dimensional score plot.

No.	Bifidobacterial strain	PC1	PC2	PC3
1	*Bifidobacterium longum* subsp. *longum* BB536	3.61	-1.88	-0.81
2	*Bifidobacterium longum* subsp. *longum* LMG 13197^{T}	4.81	-2.12	0.06
3	*Bifidobacterium longum* subsp. *infantis* LMG 11570	-2.68	-2.48	-0.16
4	*Bifidobacterium longum* subsp. *infantis* LMG 11588	-0.43	2.96	-2.07
5	*Bifidobacterium longum* subsp. *longum* LMG 11047	5.15	3.62	-0.26
6	*Bifidobacterium longum* subsp. *longum* 46	4.81	-2.12	0.06
7	*Bifidobacterium longum* subsp. *longum* LMG 13196	-0.70	-2.18	-1.42
8	*Bifidobacterium longum* subsp. *longum* NCC2705	4.81	-2.12	0.06
9	*Bifidobacterium longum* subsp. *longum* CUETM 172	4.61	-2.05	-0.27
10	*Bifidobacterium longum* subsp. *longum* PRO-16-10	5.28	-1.95	-0.06
11	*Bifidobacterium longum* subsp. *longum* CUETM 193	5.23	3.01	-0.38
12	*Bifidobacterium longum* subsp. *longum* CUETM 239	0.57	3.45	-0.94
13	*Bifidobacterium longum* subsp. *longum* CUETM 290	5.13	-2.32	0.77
14	*Bifidobacterium longum* subsp. *longum* CUETM 171	5.37	-0.67	-0.09
15	*Bifidobacterium longum* subsp. *longum* ATCC 51870	-0.70	-2.18	-1.42
16	*Bifidobacterium pseudolongum* subsp. *pseudolongum* LMG 11594	-1.42	5.01	-0.92
17	*Bifidobacterium pseudolongum* subsp. *globosum* LMG 11614	-2.60	4.34	-0.22
18	*Bifidobacterium bifidum* DSM 20082	-3.13	-2.58	0.19
19	*Bifidobacterium bifidum* LMG 11583	-3.13	-2.58	0.19
20	*Bifidobacterium bifidum* LMG 11582	-3.13	-2.58	0.19
21	*Bifidobacterium bifidum* LMG 13195	-3.13	-2.58	0.19
22	*Bifidobacterium animalis* subsp. lactis DN-173 010	-1.89	4.86	-0.55
23	*Bifidobacterium animalis* subsp. lactis R-17143	-0.96	5.14	-1.03
24	*Bifidobacterium animalis* subsp. *lactis* BB-12	0.07	5.11	-1.81
25	*Bifidobacterium dentium* LMG 10507	-0.65	-1.74	-1.73
26	*Bifidobacterium breve* LMG 13194	-2.88	-2.21	0.21
27	*Bifidobacterium breve* LMG 13208	-3.13	-2.58	0.19
28	*Bifidobacterium breve* LMG 11040	-3.13	-2.58	0.19
29	*Bifidobacterium breve* Yakult	-3.13	-2.58	0.19
30	*Bifidobacterium thermophilum* LMG 11574	-3.08	-2.14	-0.12
31	*Bifidobacterium catenulatum* LMG 11043^{T}	-0.14	2.16	12.94

32	*Bifidobacterium gallicum* LMG 11596^{T}	-2.19	3.84	-0.32
33	*Bifidobacterium angulatum* LMG 11039^{T}	-0.60	2.89	-0.42
34	*Bifidobacterium angulatum* LMG 11568	-1.74	-2.18	-0.87
35	*Bifidobacterium adolescentis* LMG 10734	-3.25	0.42	-0.09
36	*Bifidobacterium adolescentis* LMG 10502^{T}	-1.65	1.59	0.54

Figure S1. Carbohydrate degradation by bifidobacterial strains belonging to cluster I.

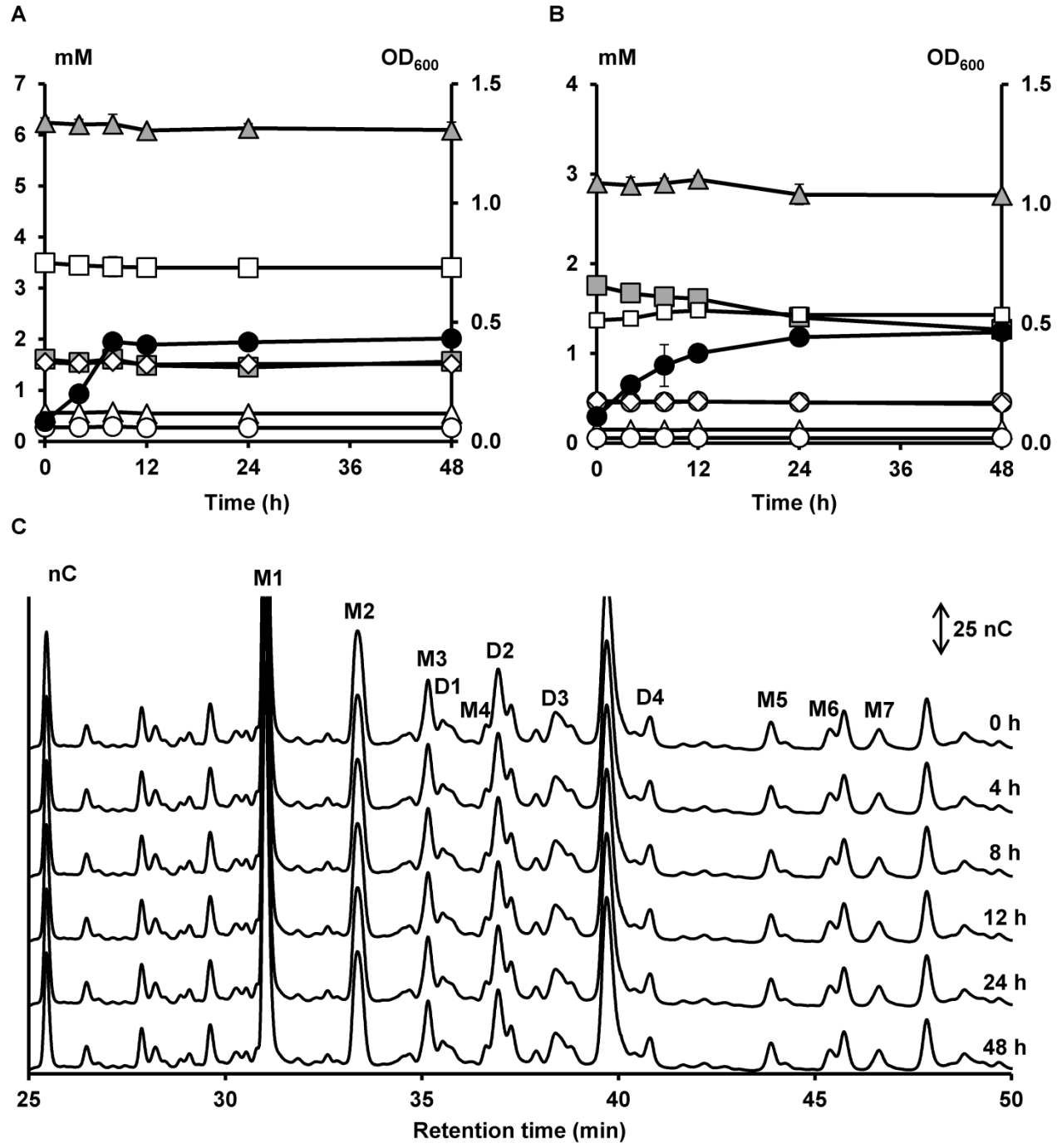

Figure S1.A. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *infantis* LMG 11570 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Growth is represented as OD_{600} measurements (●). Monosaccharide, XOS, and $XOS_{(A)XOS}$ consumption are represented as

residual concentrations of arabinose (●), xylose (■), xylobiose (▲), xylotriose (□), xylotetraose (◊), xylopentaose (Δ), and xylohexaose (○) in mM. (C) represents an enlargement of the AXOS HPAEC-PAD chromatograms between 25.0 and 50.0 min of elution. D1-D4, AXOS peaks containing at least one doubly substituted Xyl*p* residue. M1-M7, AXOS peaks containing at least one monosubstituted Xyl*p* residue.

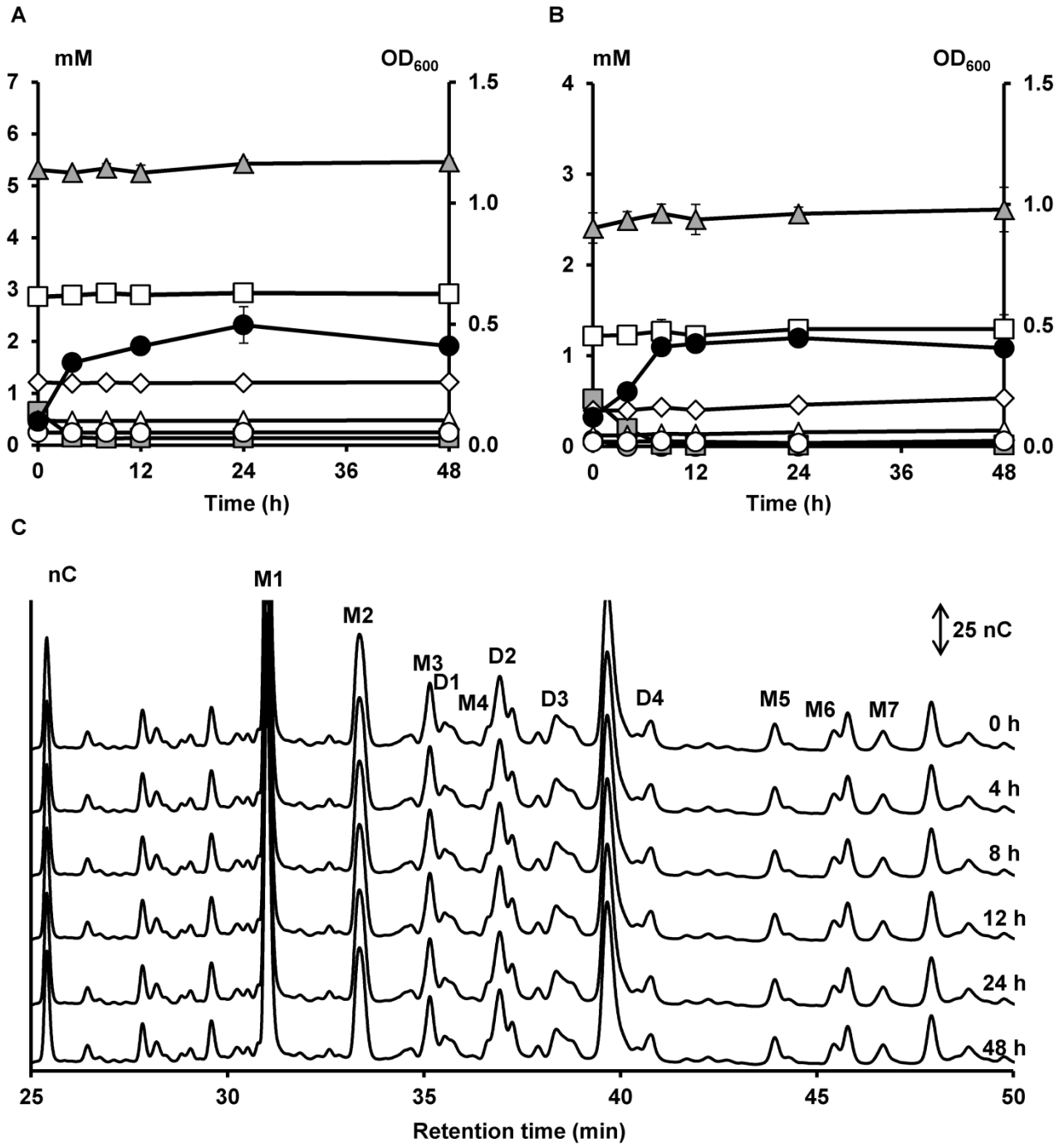

Figure S1.B. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* LMG 13196 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

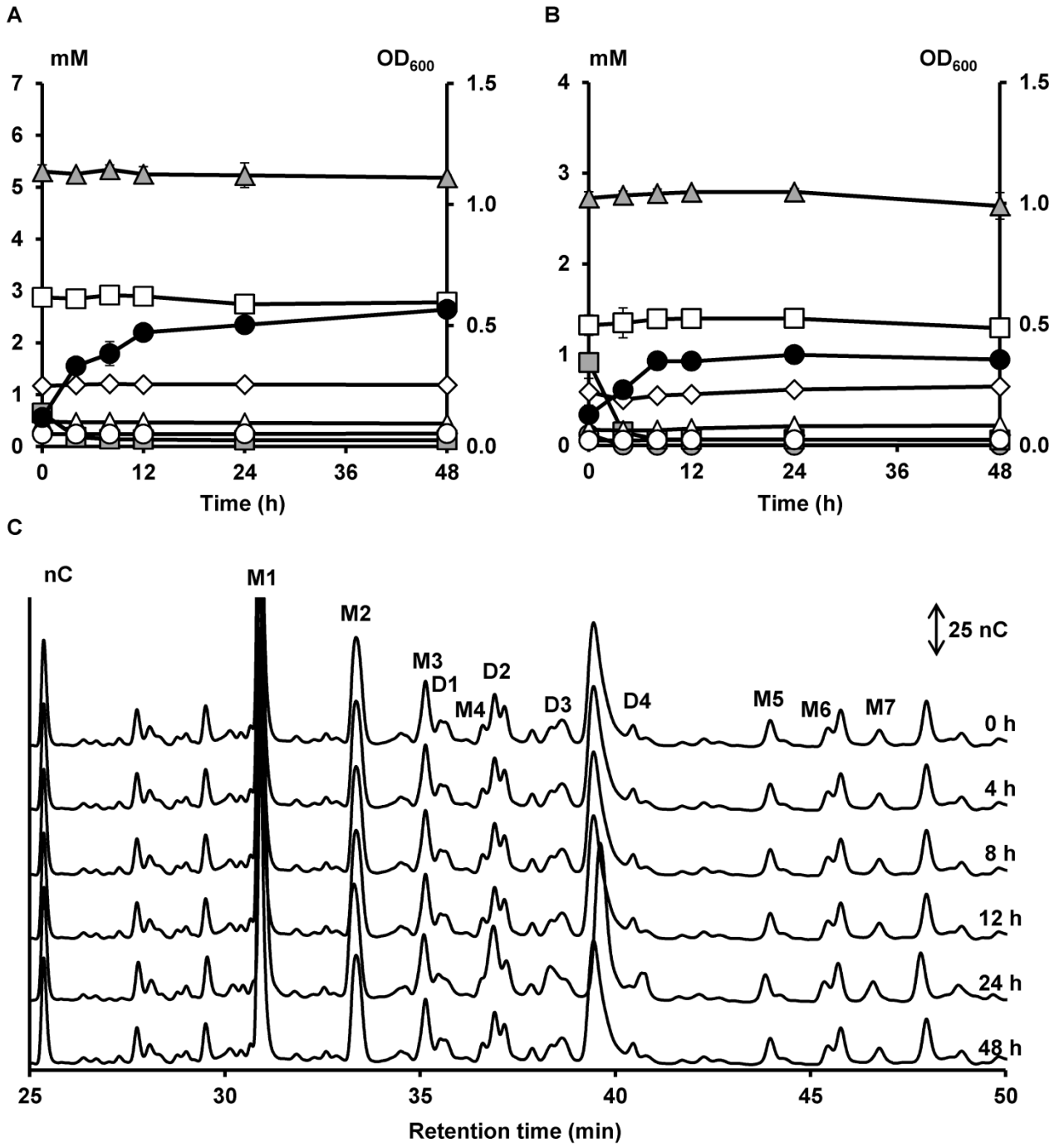

Figure S1.C. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* ATCC 51870 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

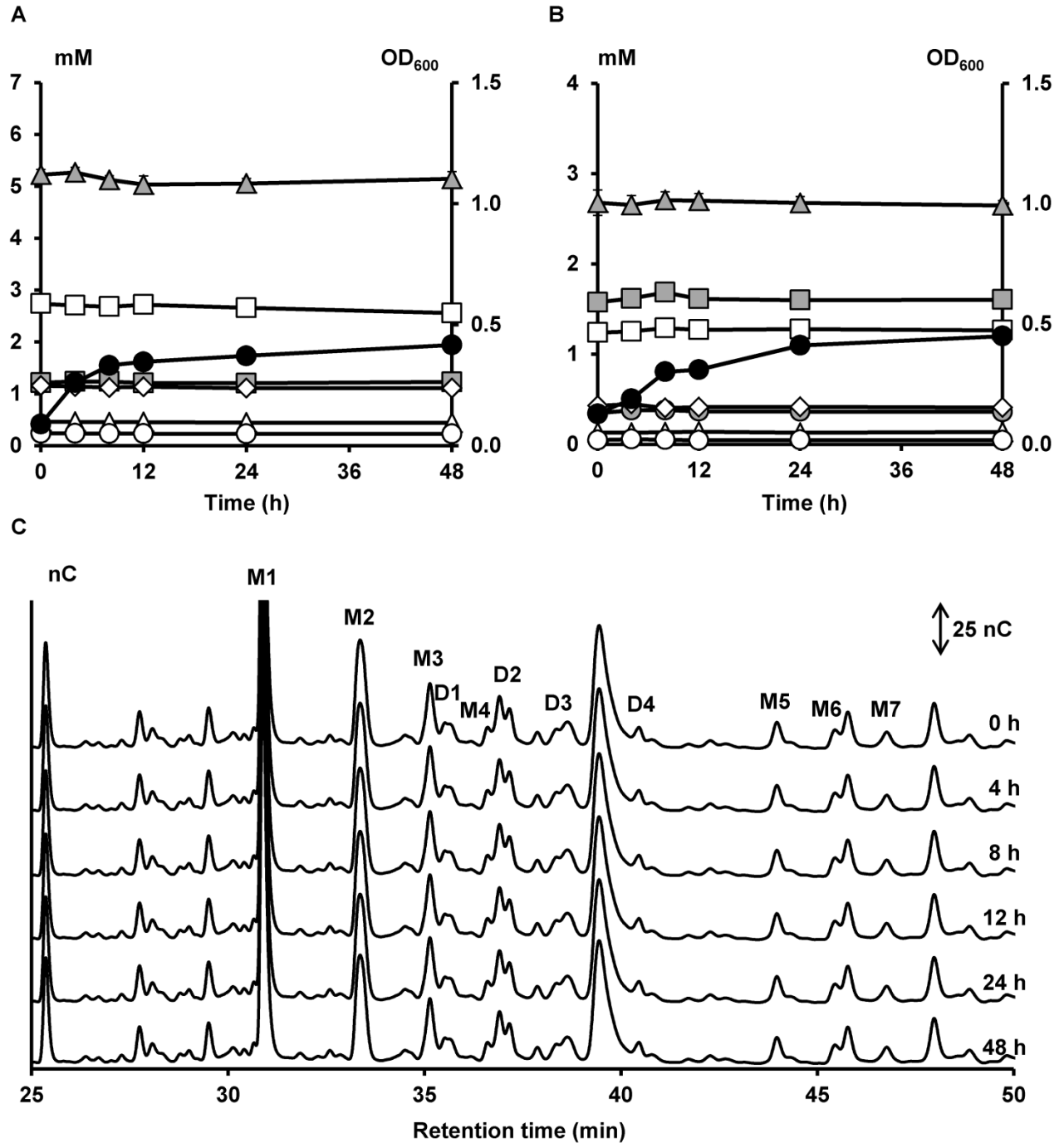

Figure S1.D. Growth of and carbohydrate degradation by *Bifidobacterium bifidum* DSM 20082 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

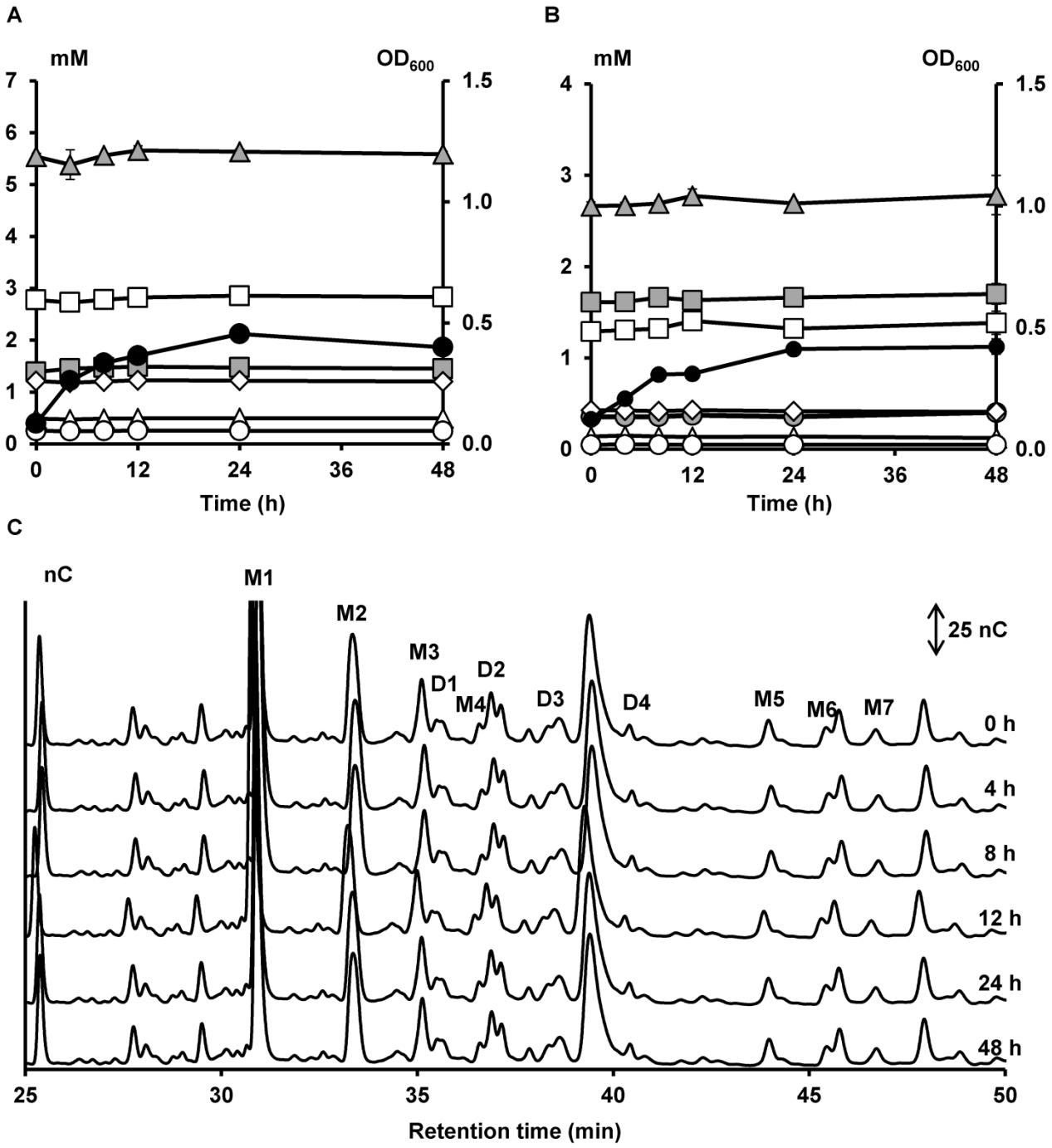

Figure S1.E. Growth of and carbohydrate degradation by *Bifidobacterium bifidum* LMG 11582 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

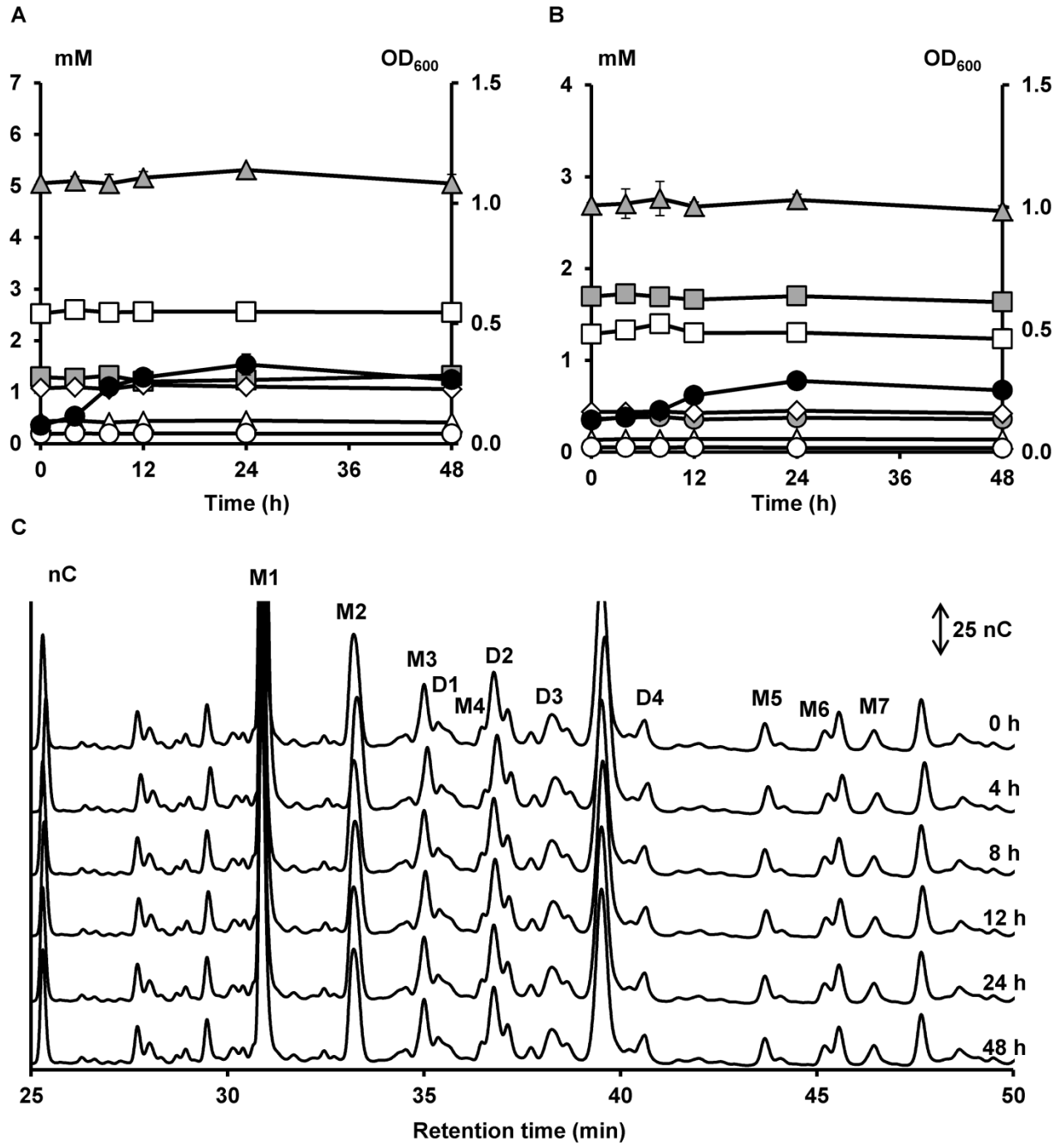

Figure S1.F. Growth of and carbohydrate degradation by *Bifidobacterium bifidum* LMG 13195 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

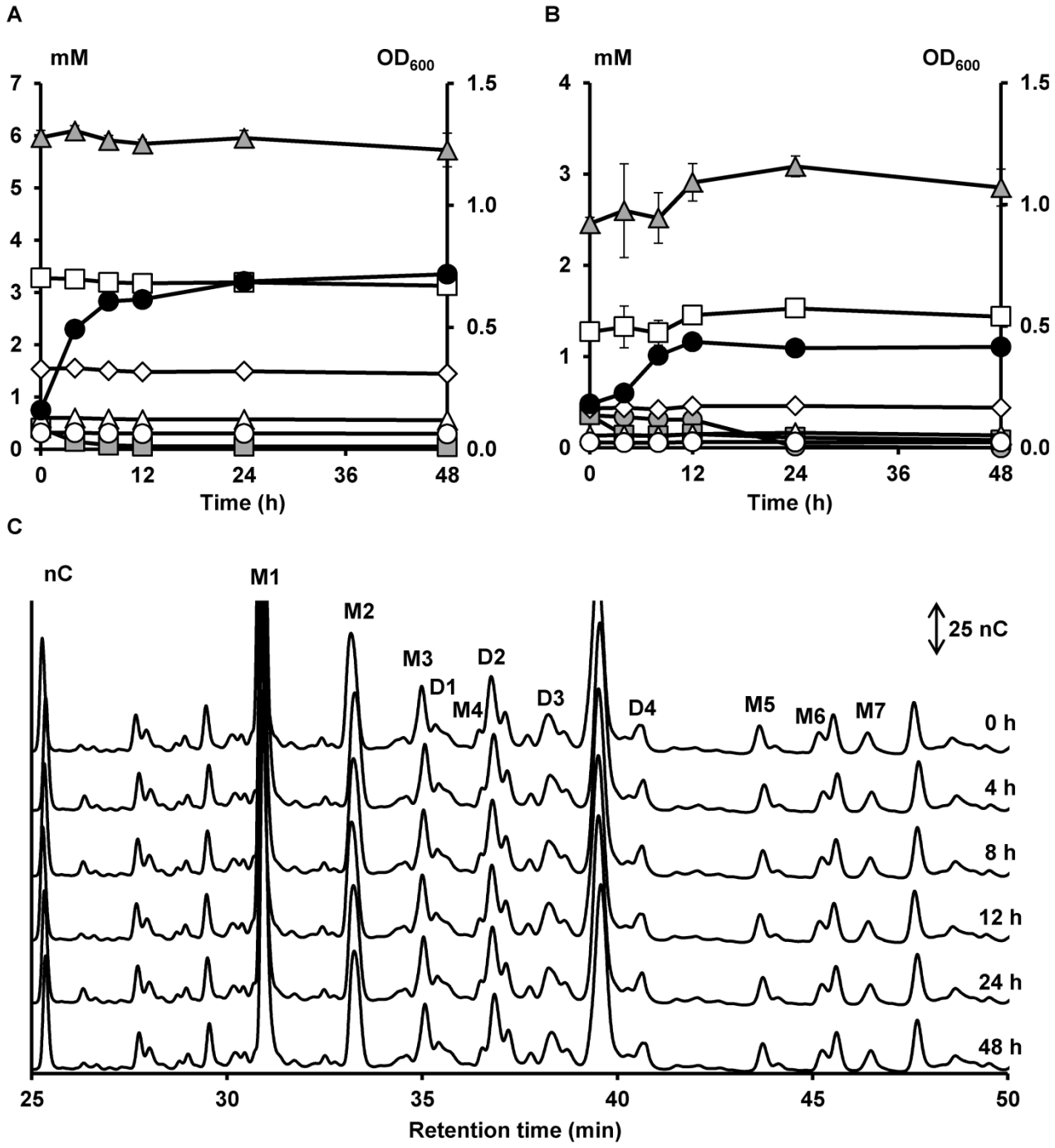

Figure S1.G. Growth of and carbohydrate degradation by *Bifidobacterium dentium* LMG 10507 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

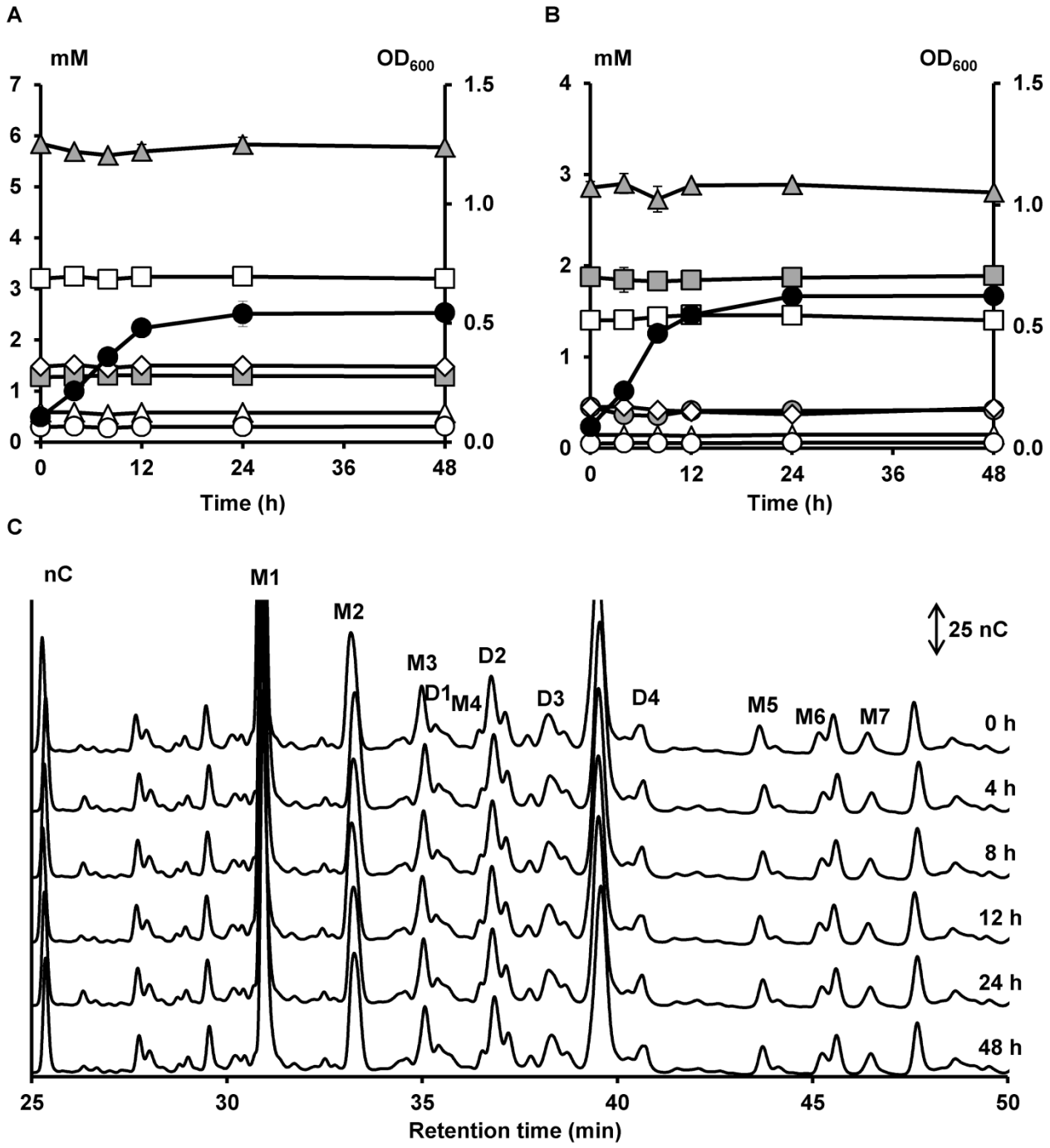

Figure S1.H. Growth of and carbohydrate degradation by *Bifidobacterium breve* LMG 13194 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

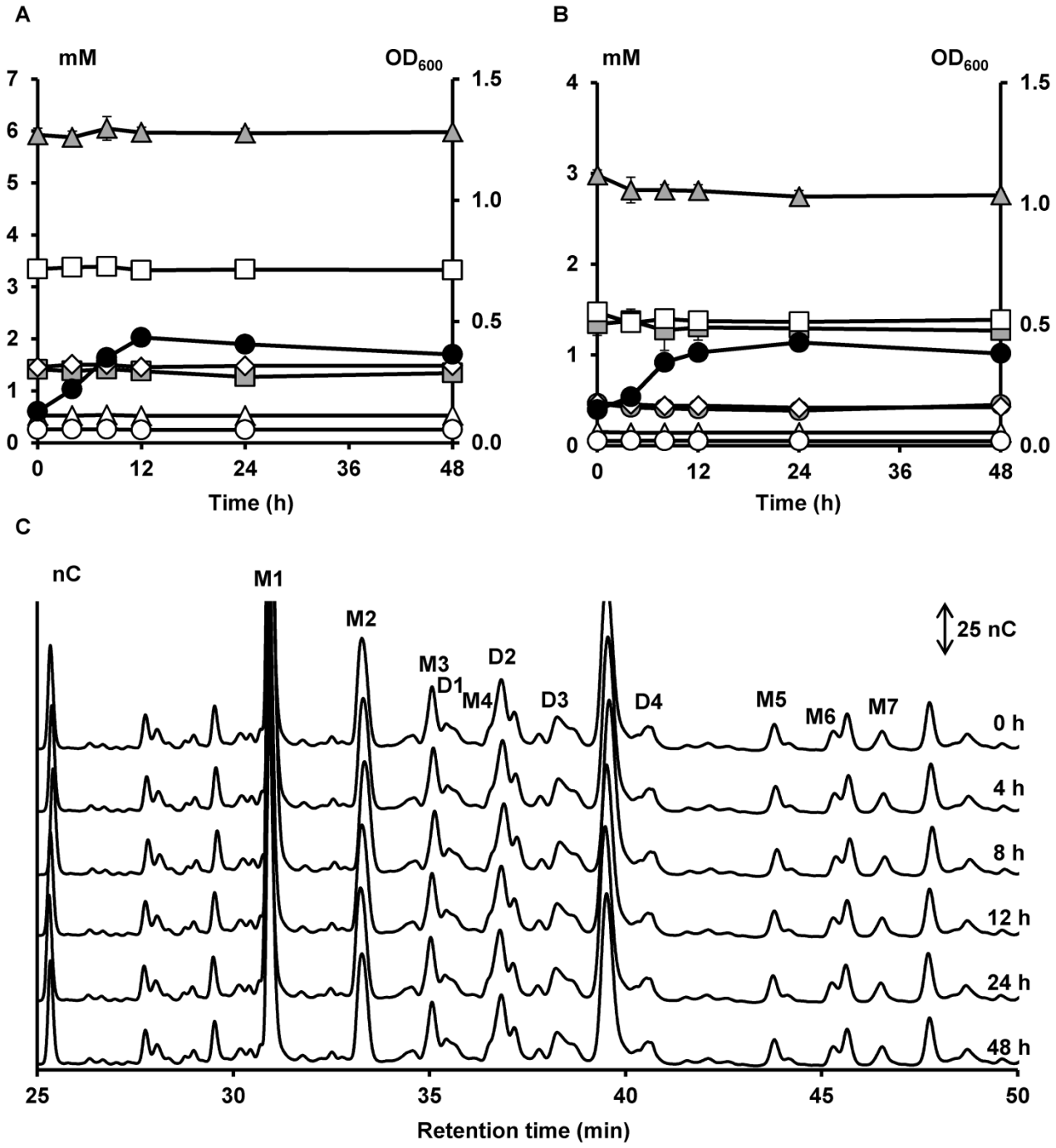

Figure S1.I. Growth of and carbohydrate degradation by *Bifidobacterium breve* LMG 13208 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

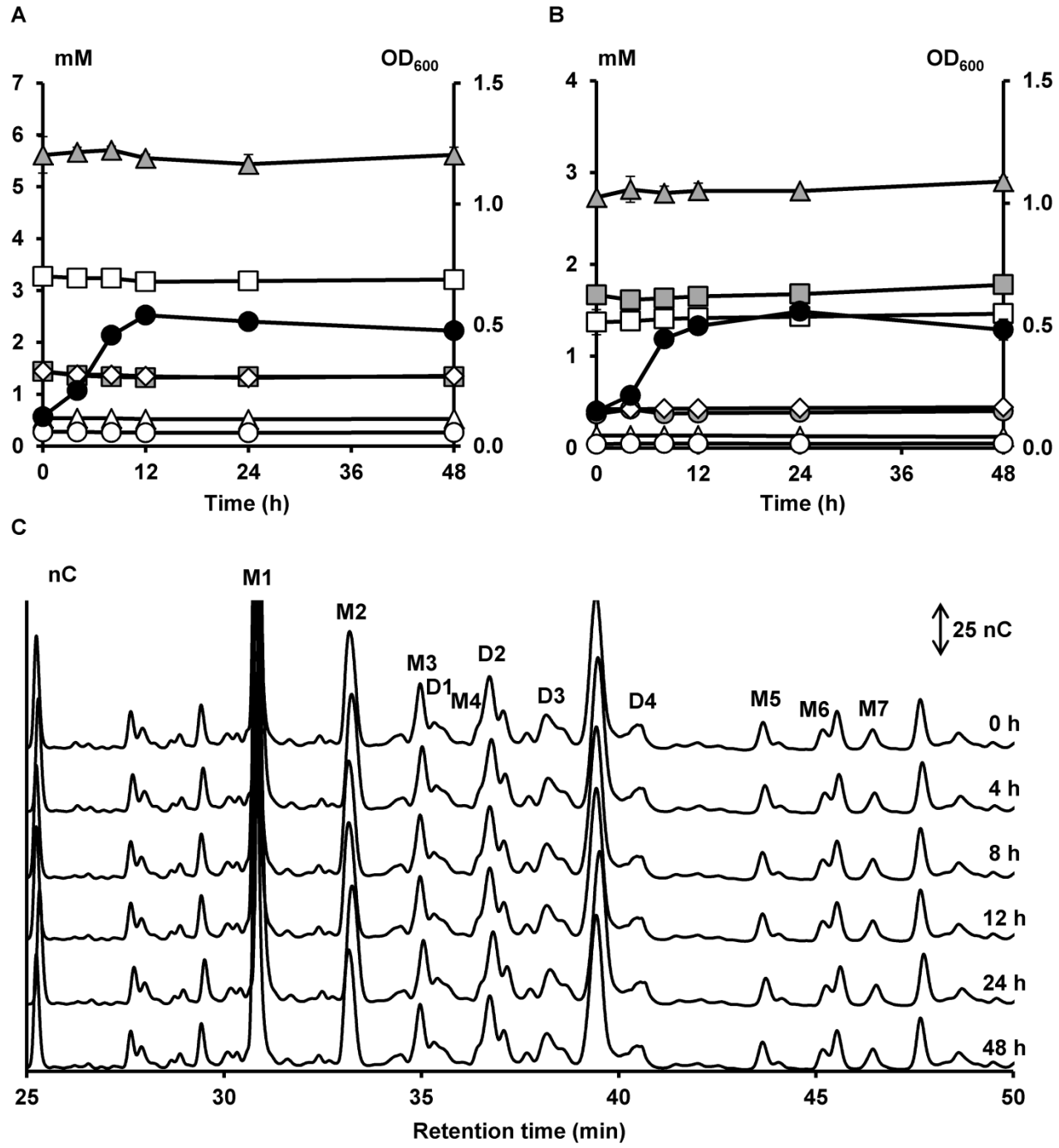

Figure S1.J. Growth of and carbohydrate degradation by *Bifidobacterium breve* LMG 11040 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

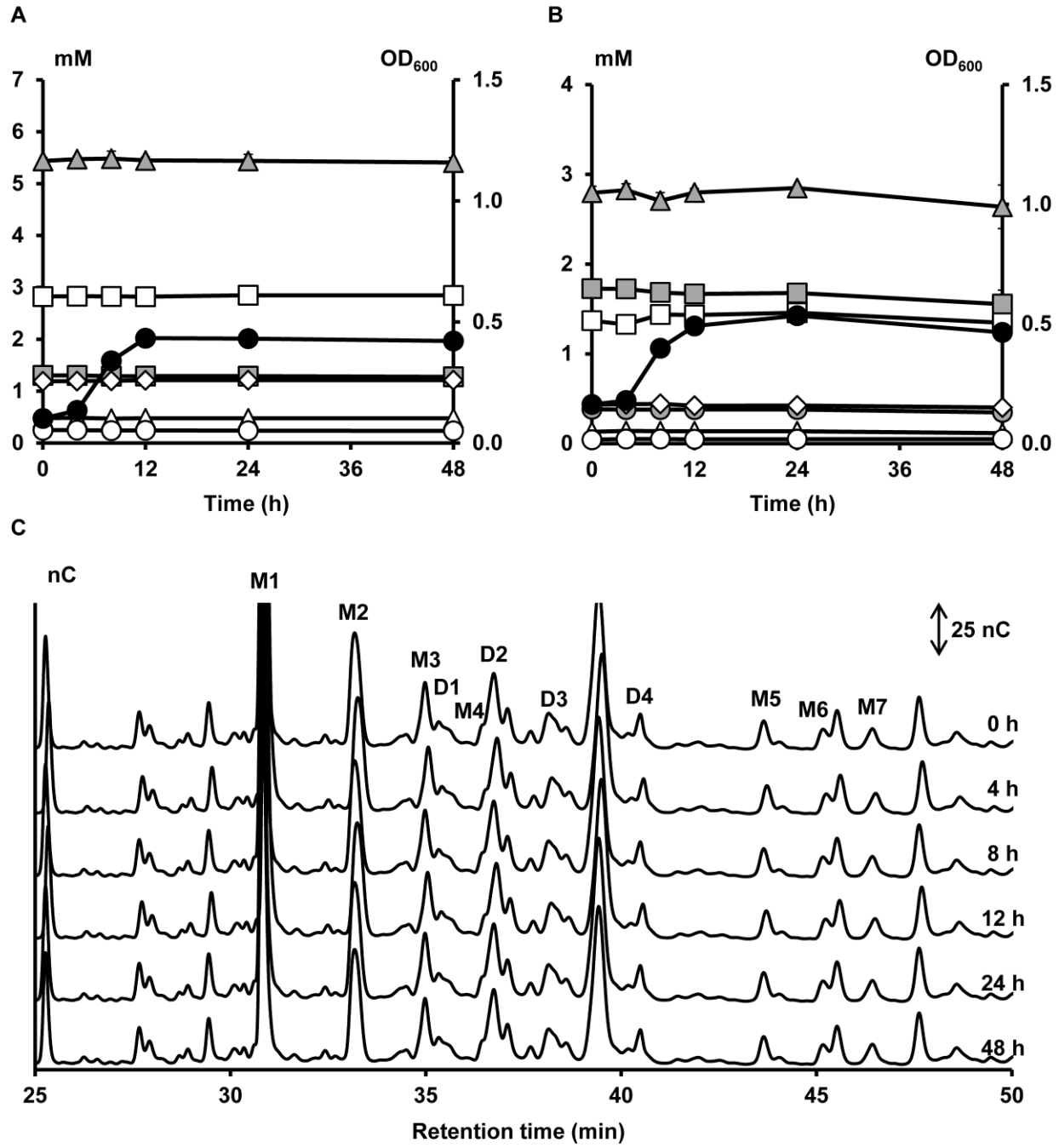

Figure S1.K. Growth of and carbohydrate degradation by *Bifidobacterium breve* Yakult in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

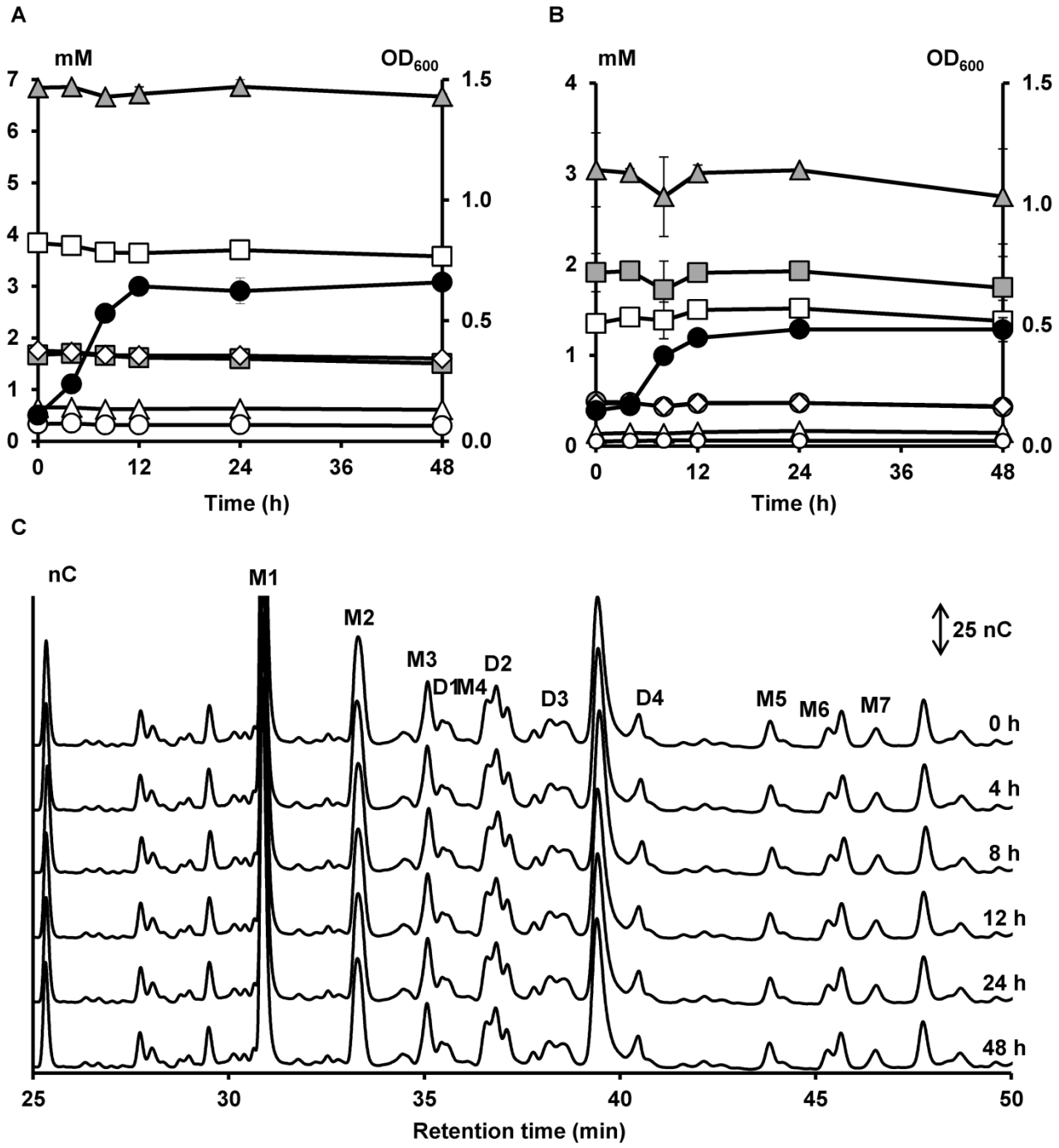

Figure S1.L. Growth of and carbohydrate degradation by *Bifidobacterium thermophilum* LMG 11574 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

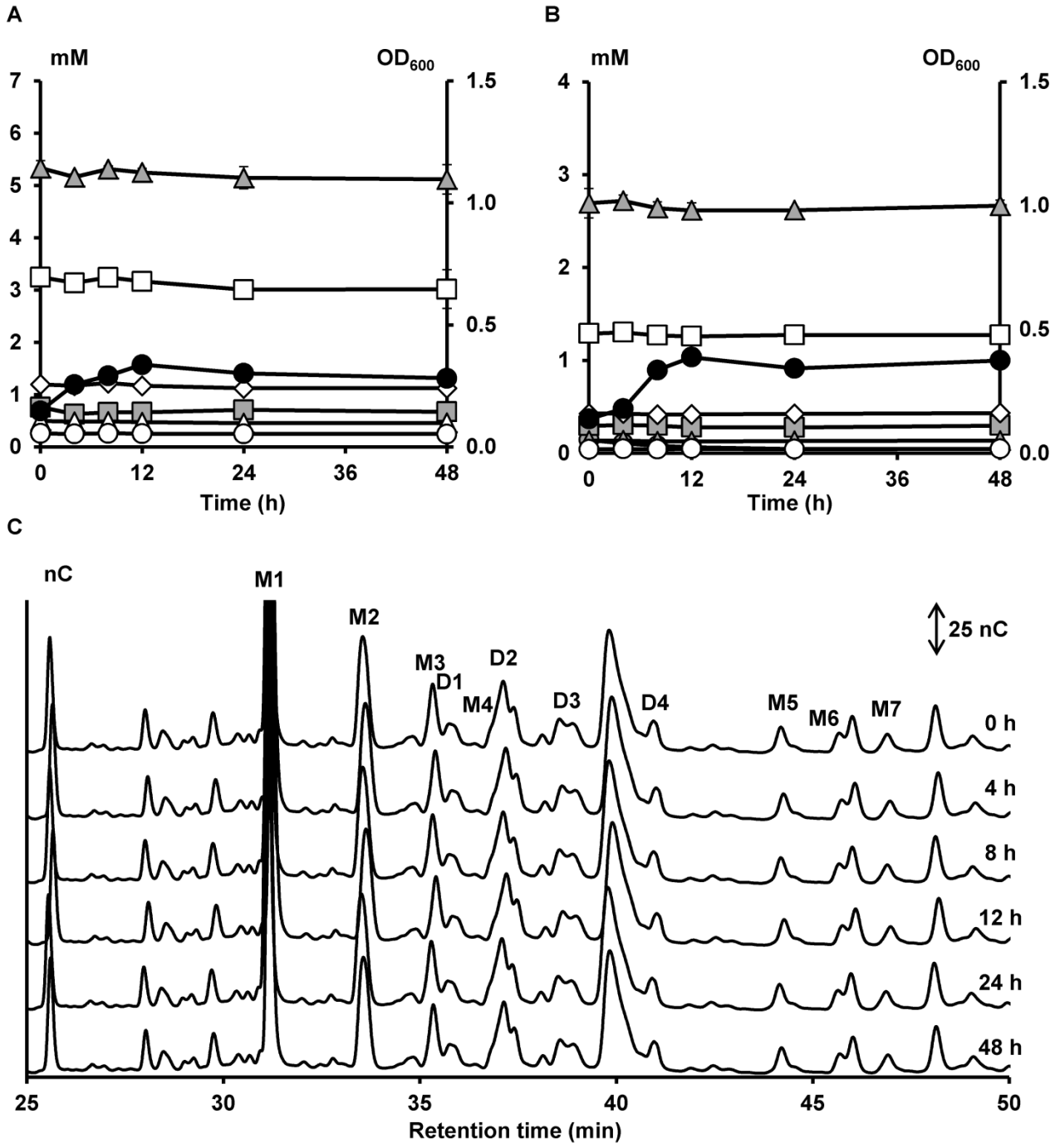

Figure S1.M. Growth of and carbohydrate degradation by *Bifidobacterium angulatum* LMG 11568 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

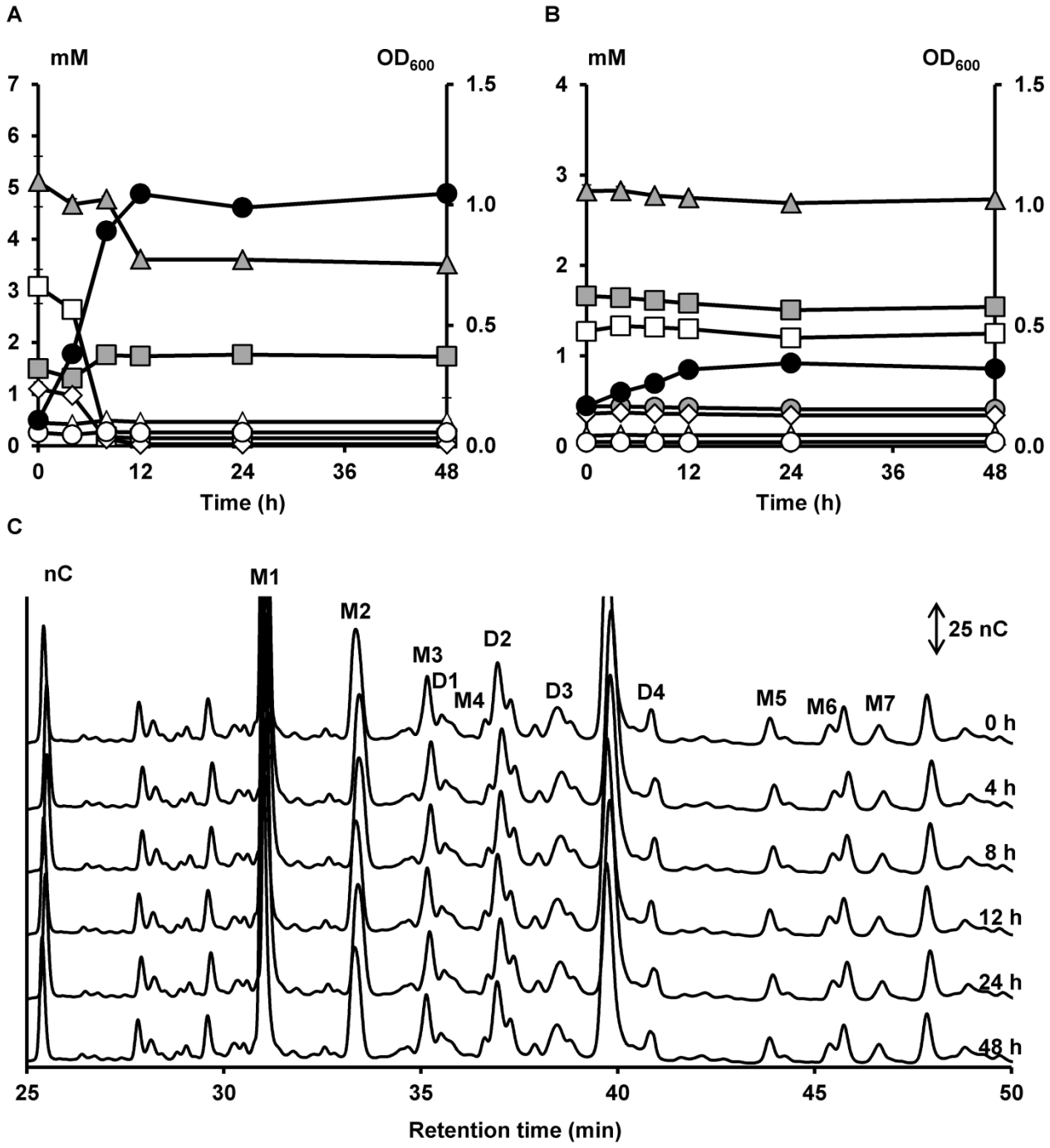

Figure S1.N. Growth of and carbohydrate degradation by *Bifidobacterium adolescentis* LMG 10734 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S1.A.

Figure S2. Carbohydrate degradation by bifidobacterial strains belonging to cluster II.

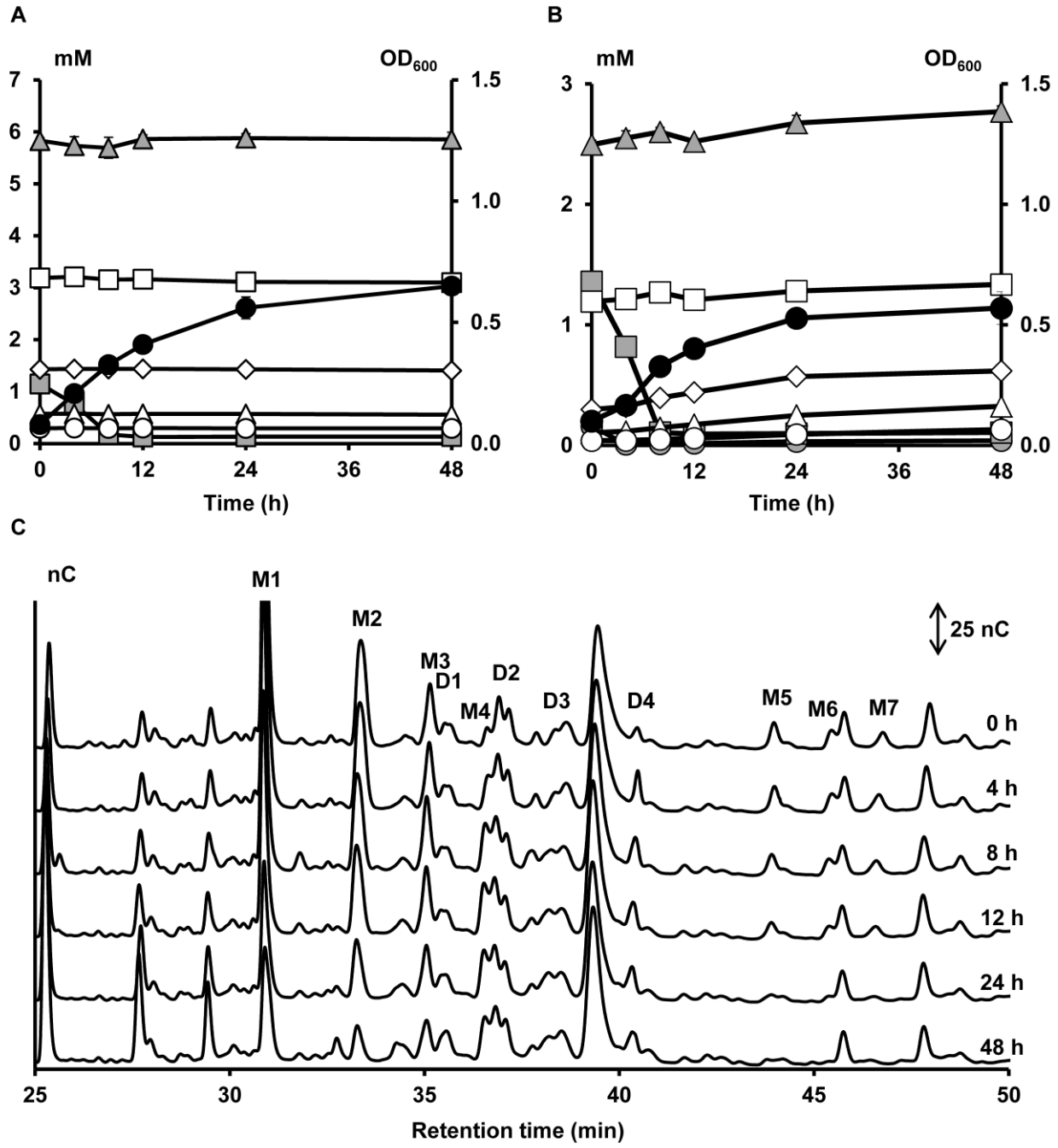

Figure S2.A. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* BB536 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Growth is represented as OD_{600} measurements (●). Monosaccharide, XOS, and $XOS_{(A)XOS}$ consumption are represented as

residual concentrations of arabinose (●), xylose (■), xylobiose (▲), xylotriose (□), xylotetraose (◊), xylopentaose (Δ), and xylohexaose (○) in mM. (C) represents an enlargement of the AXOS HPAEC-PAD chromatograms between 25.0 and 50.0 min of elution. D1-D4, AXOS peaks containing at least one doubly substituted Xyl*p* residue. M1-M7, AXOS peaks containing at least one monosubstituted Xyl*p* residue.

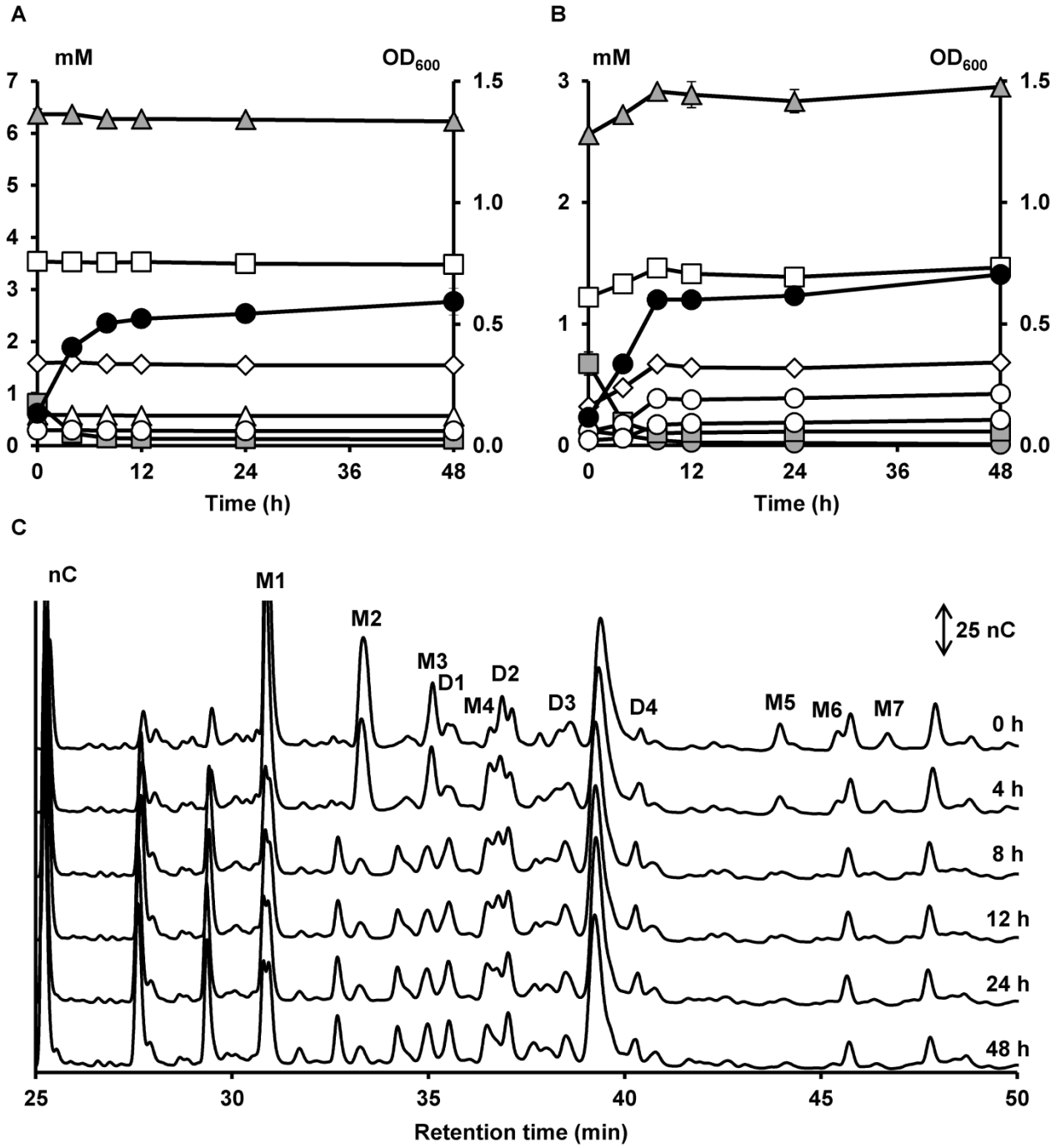

Figure S2.B. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* LMG 13197^T in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S2.A.

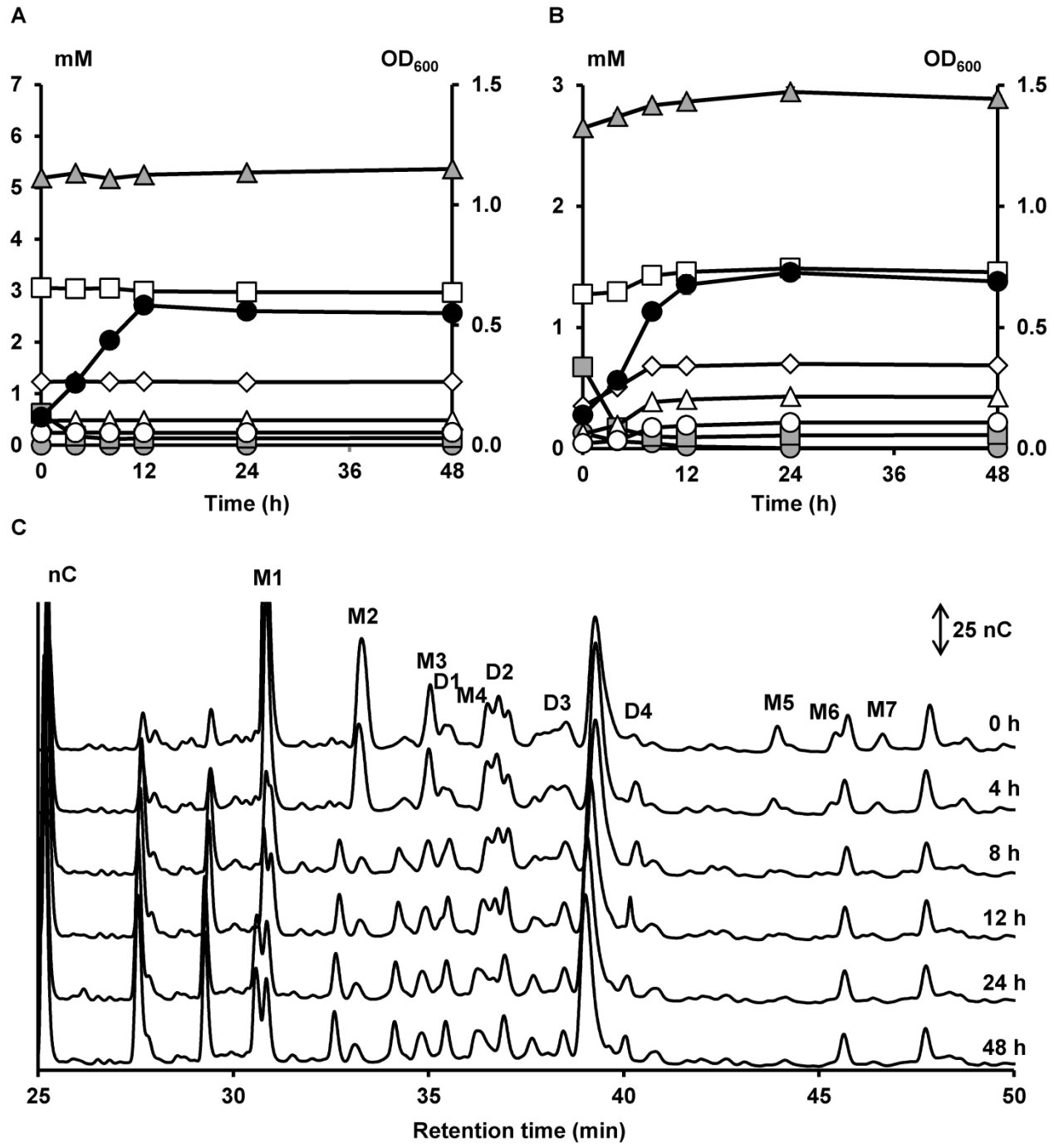

Figure S2.C. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* 46 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S2.A.

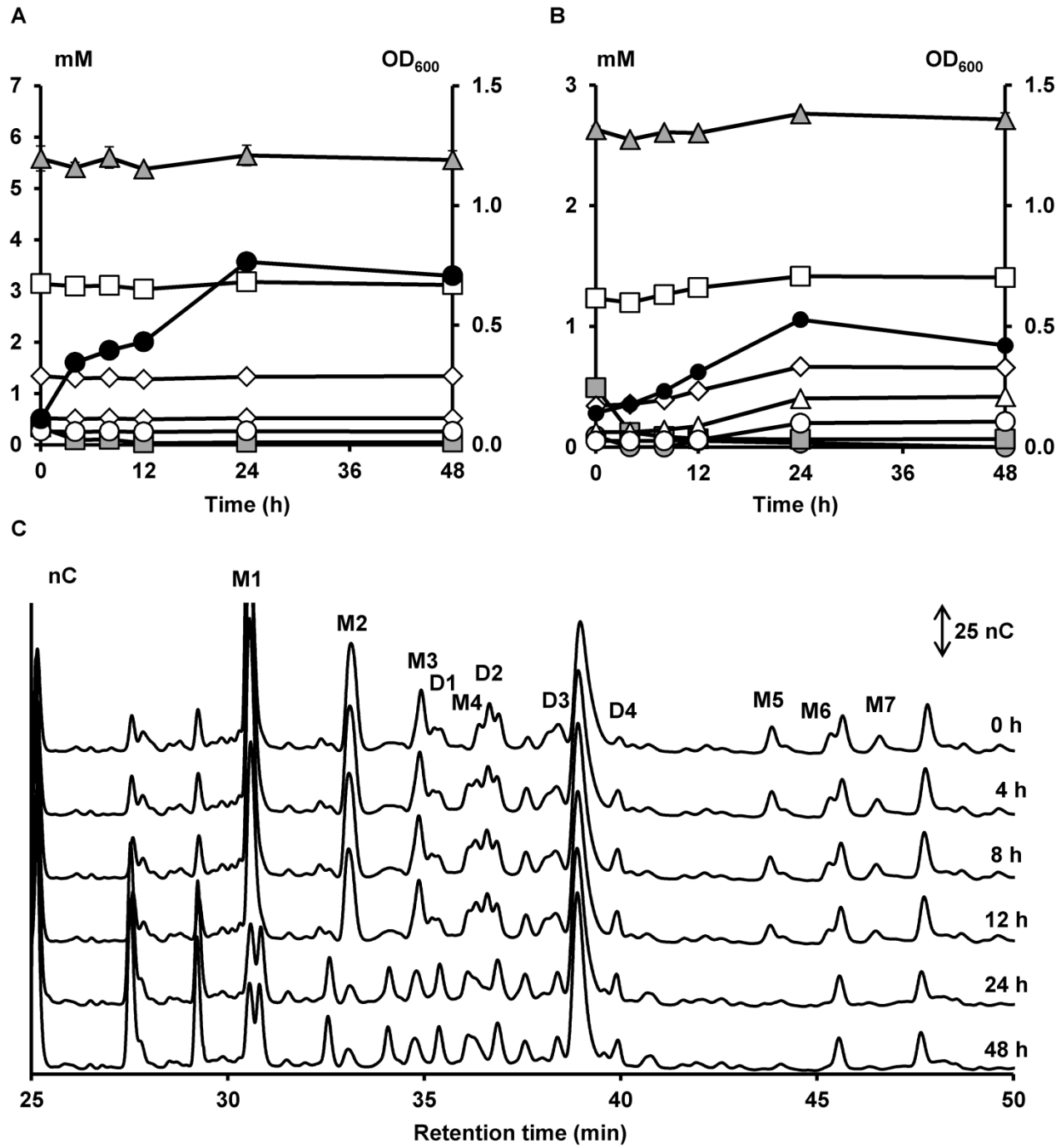

Figure S2.D. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* CUETM 172 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S2.A.

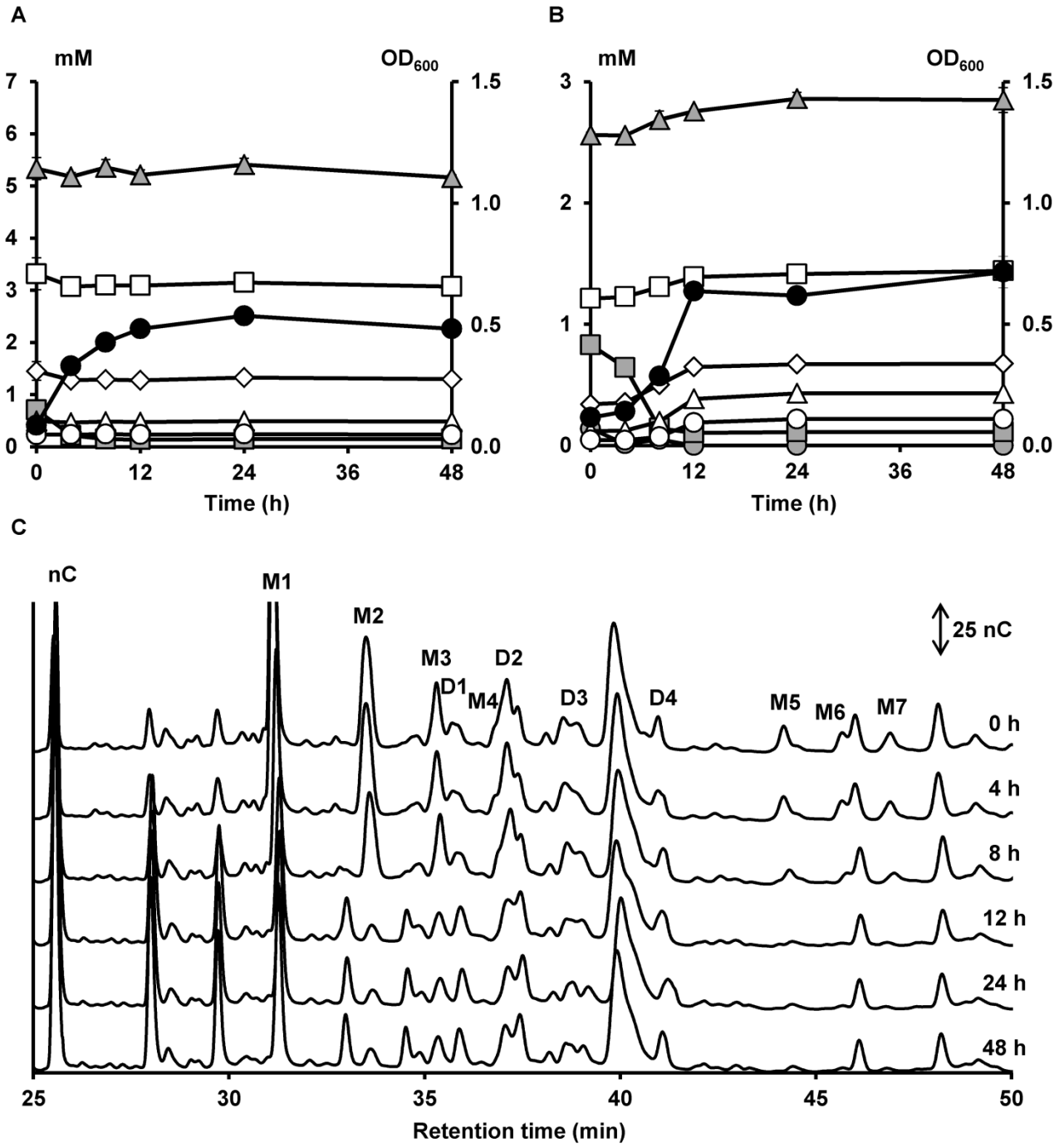

Figure S2.E. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* PRO-16-10 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S2.A.

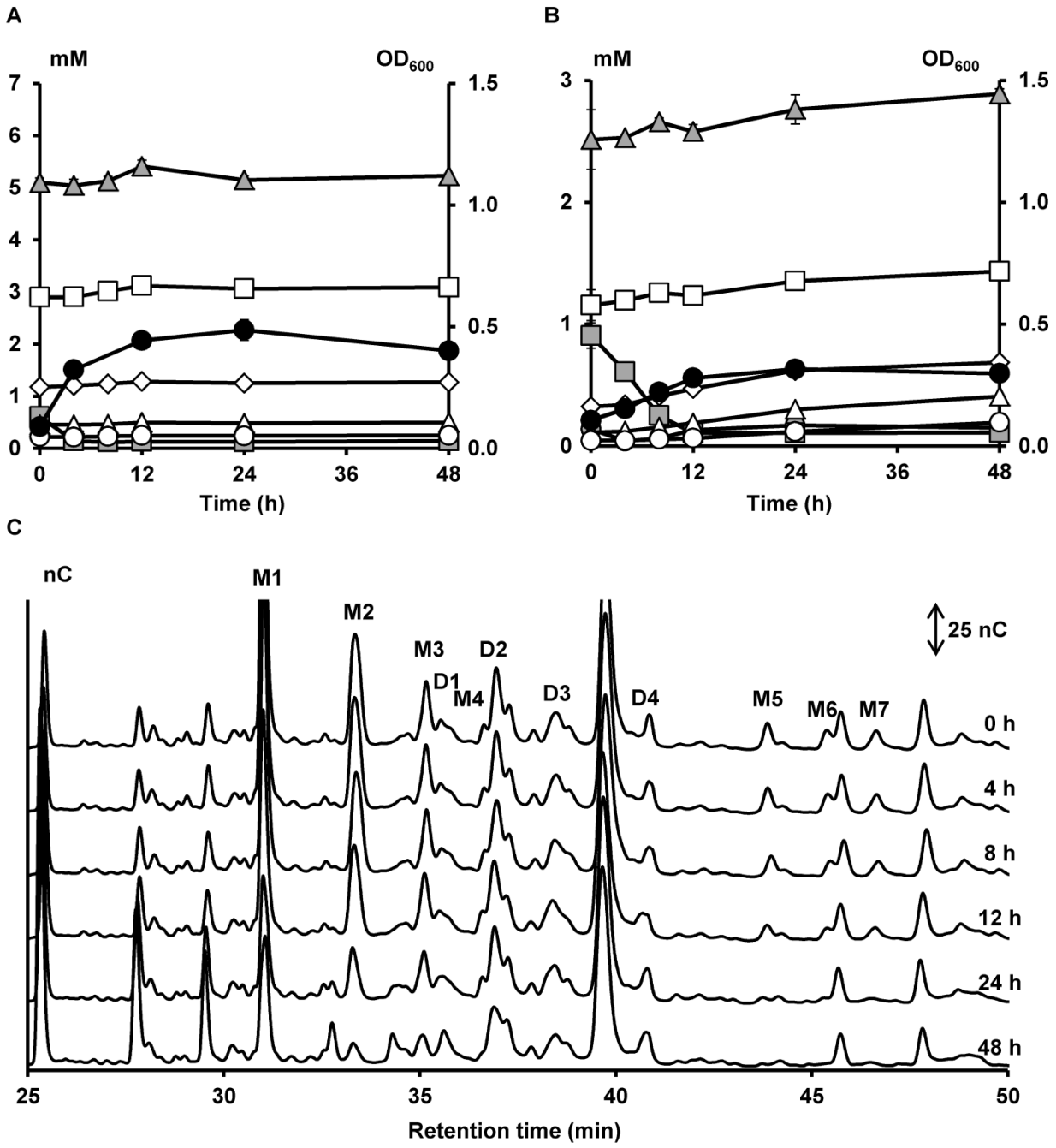

Figure S2.F. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* CUETM 290 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S2.A.

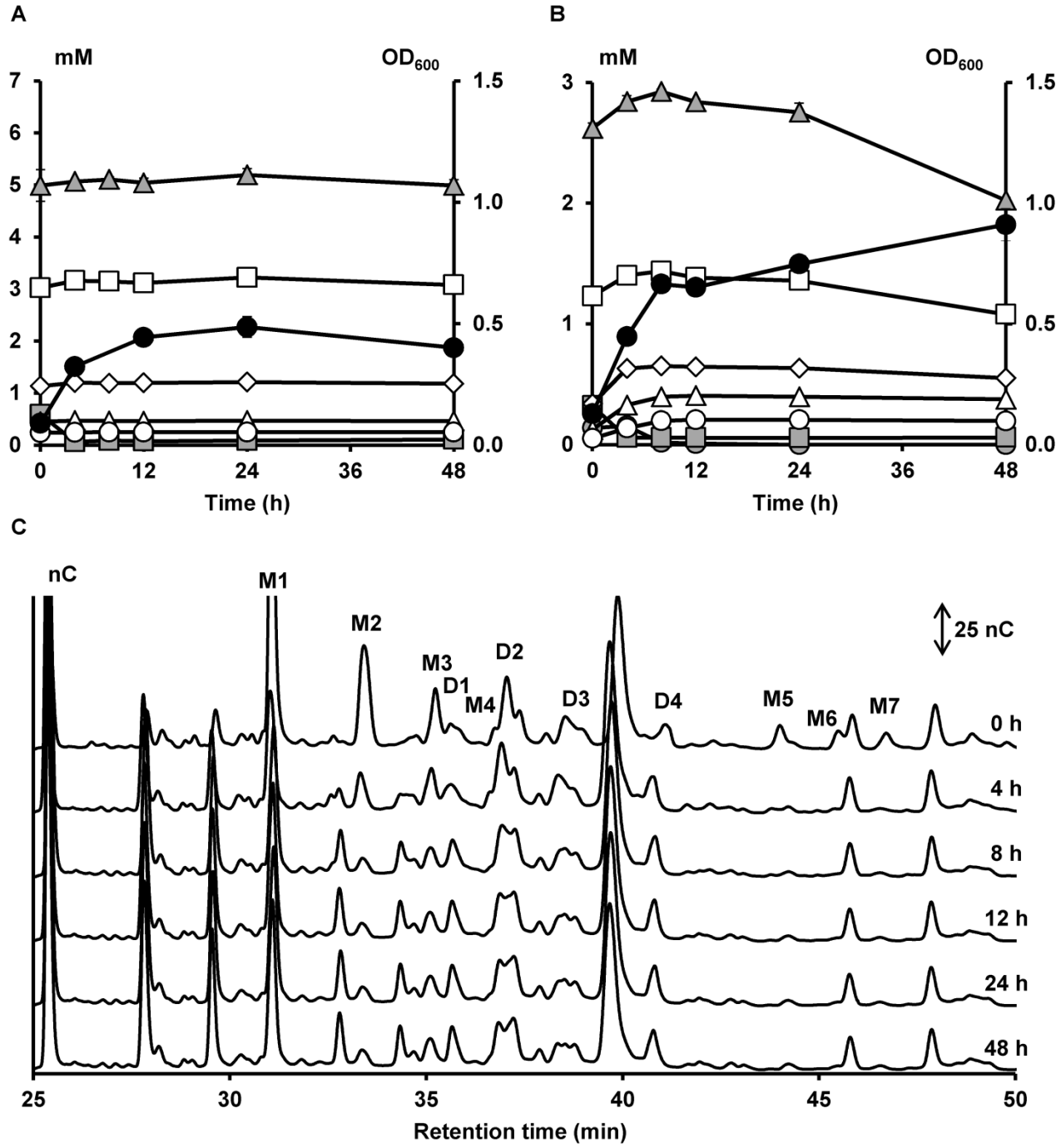

Figure S2.G. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* CUETM 171 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S2.A.

Figures S3. Carbohydrate degradation by bifidobacterial strains belonging to cluster III.

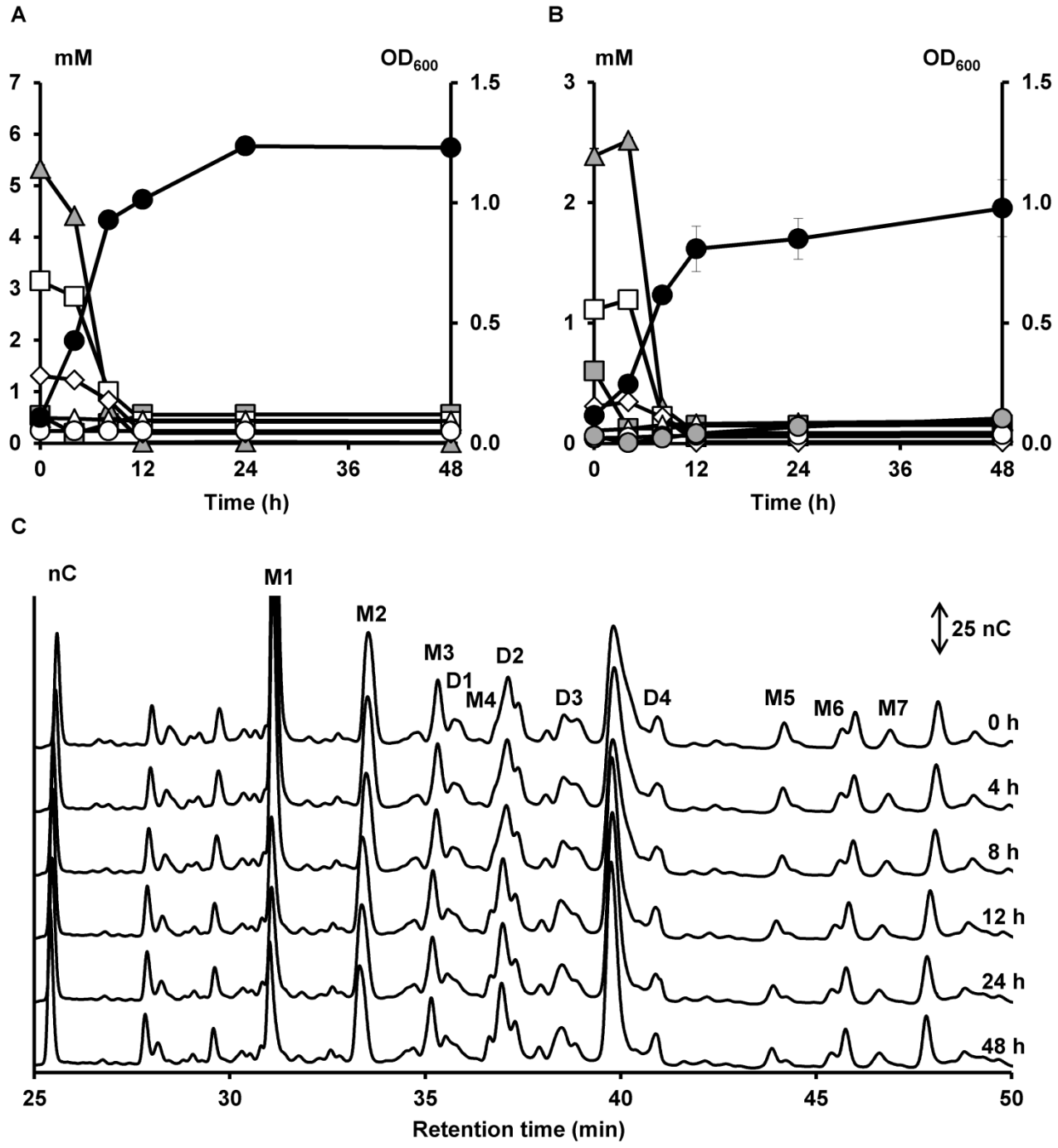

Figure S3.A. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* CUETM 239 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Growth is represented as OD_{600} measurements (●). Monosaccharide, XOS, and $XOS_{(A)XOS}$ consumption are represented as

residual concentrations of arabinose (●), xylose (■), xylobiose (▲), xylotriose (□), xylotetraose (◊), xylopentaose (Δ), and xylohexaose (○) in mM. (C) represents an enlargement of the AXOS HPAEC-PAD chromatograms between 25.0 and 50.0 min of elution. D1-D4, AXOS peaks containing at least one doubly substituted Xyl*p* residue. M1-M7, AXOS peaks containing at least one monosubstituted Xyl*p* residue.

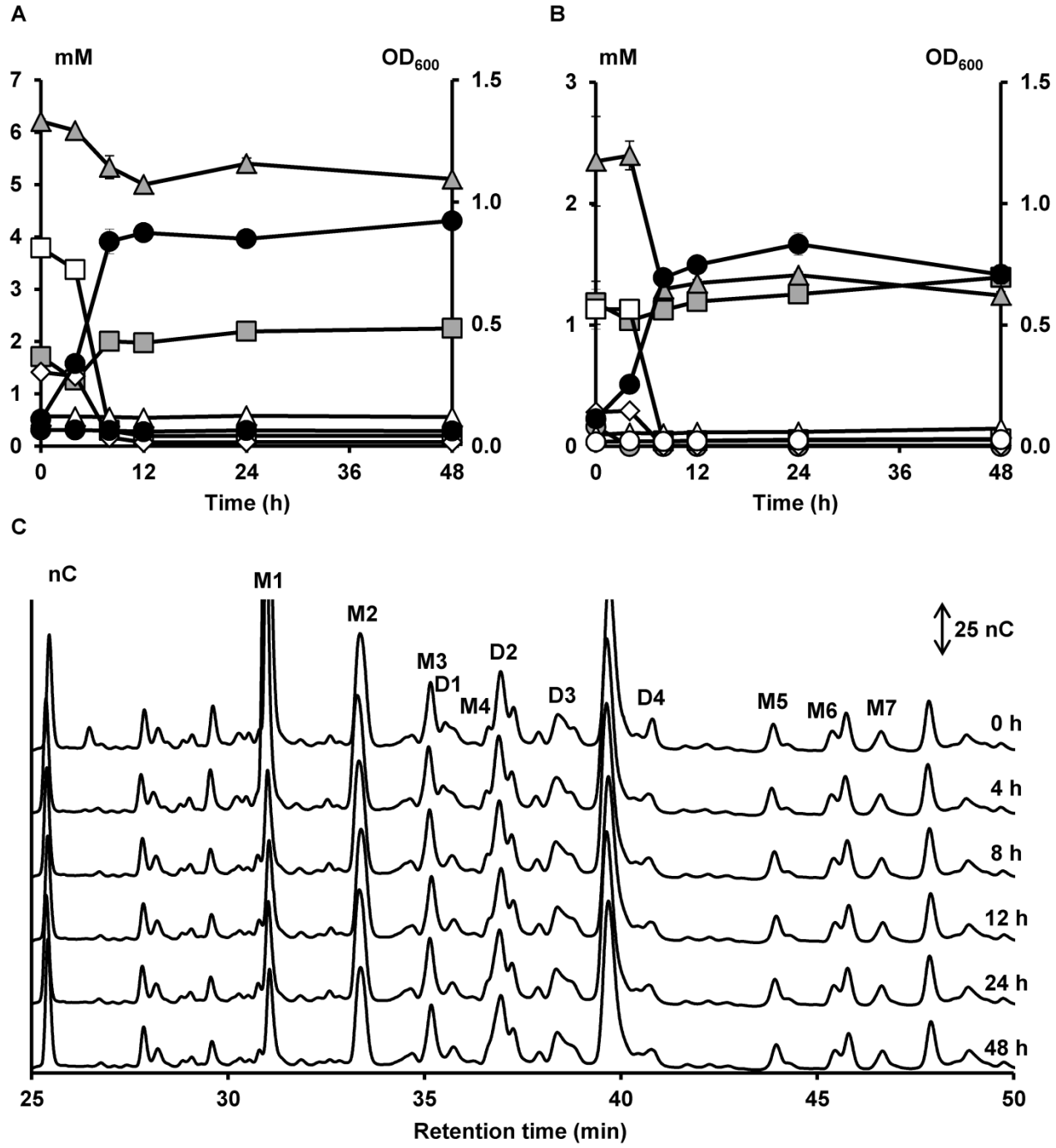

Figure S3.B. Growth of and carbohydrate degradation by *Bifidobacterium pseudolongum* subsp. *pseudolongum* LMG 11594 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

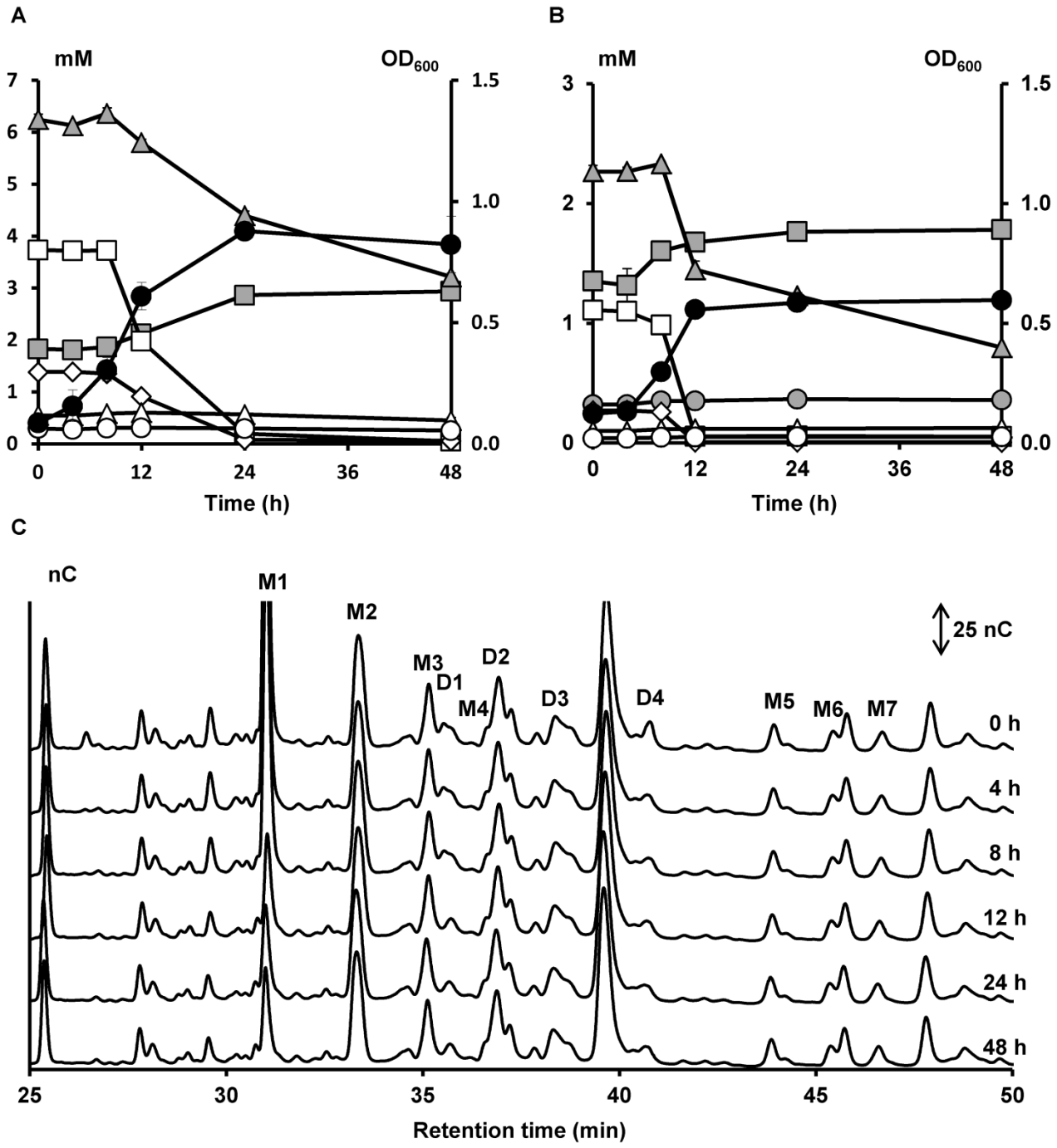

Figure S3.C. Growth of and carbohydrate degradation by *Bifidobacterium pseudolongum* subsp. *globosum* LMG 11614 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

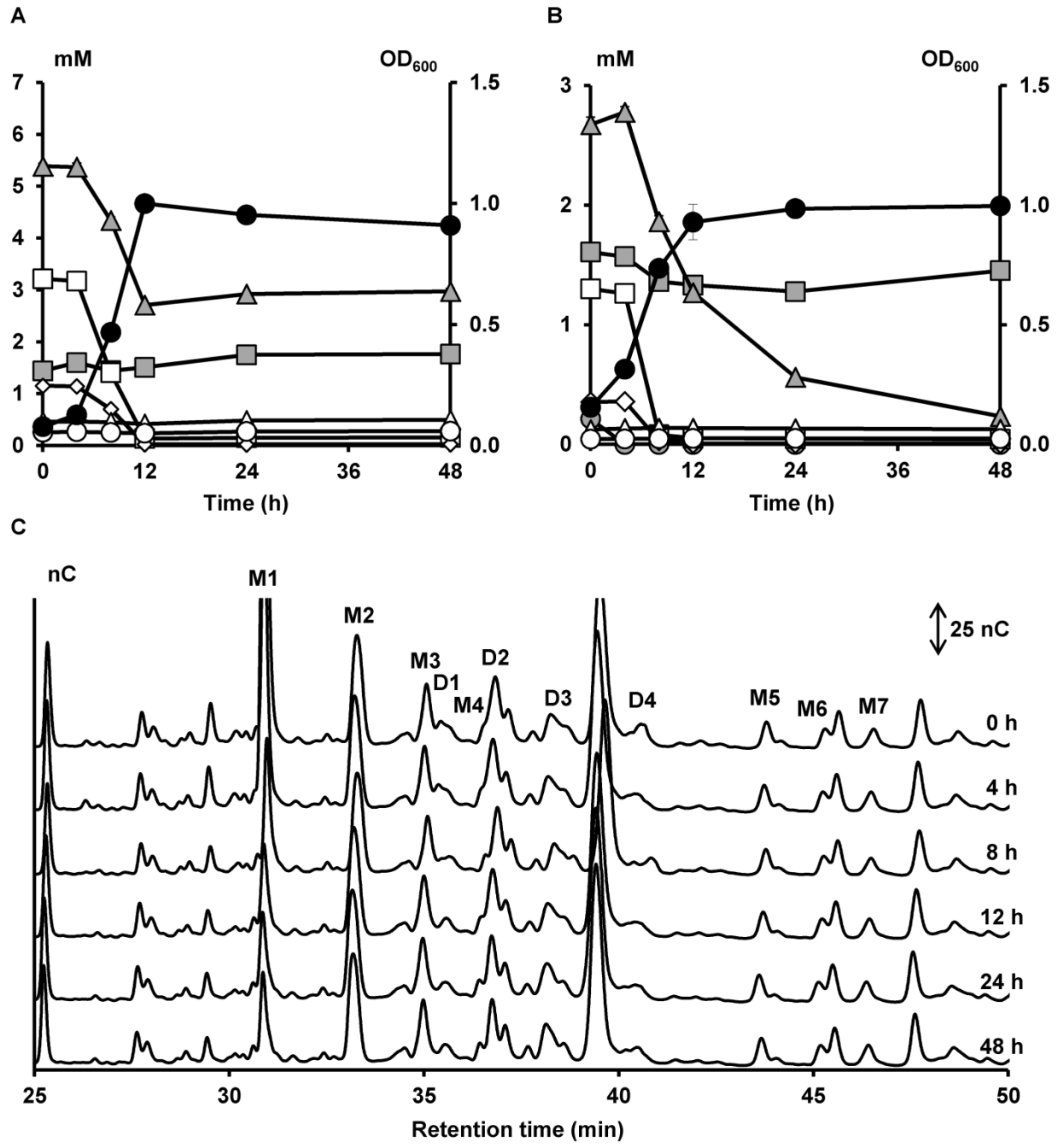

Figure S3.D. Growth of and carbohydrate degradation by *Bifidobacterium animalis* subsp. *lactis* DN-173 010 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

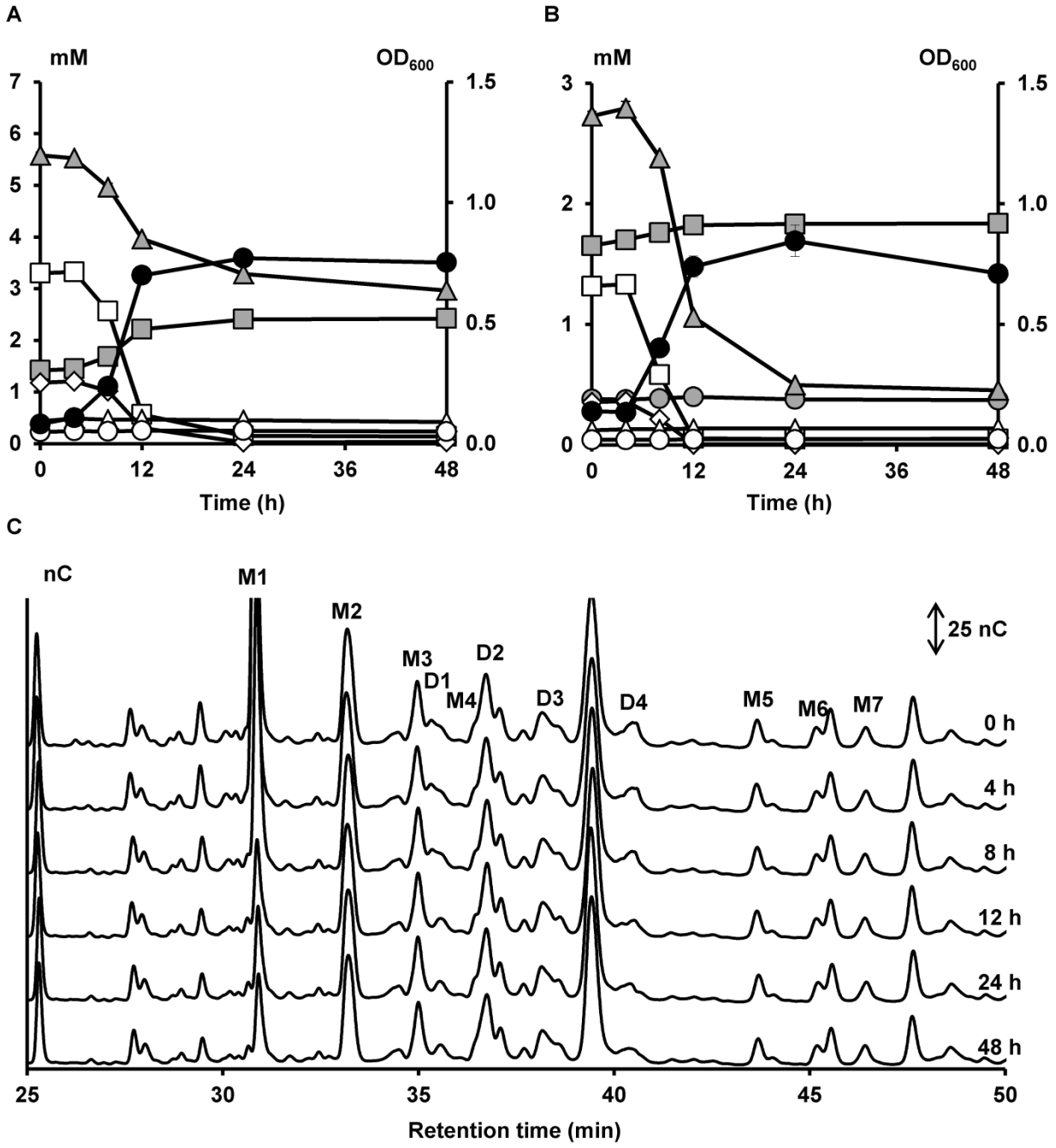

Figure S3.E. Growth of and carbohydrate degradation by *Bifidobacterium animalis* subsp. *lactis* R-17143 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

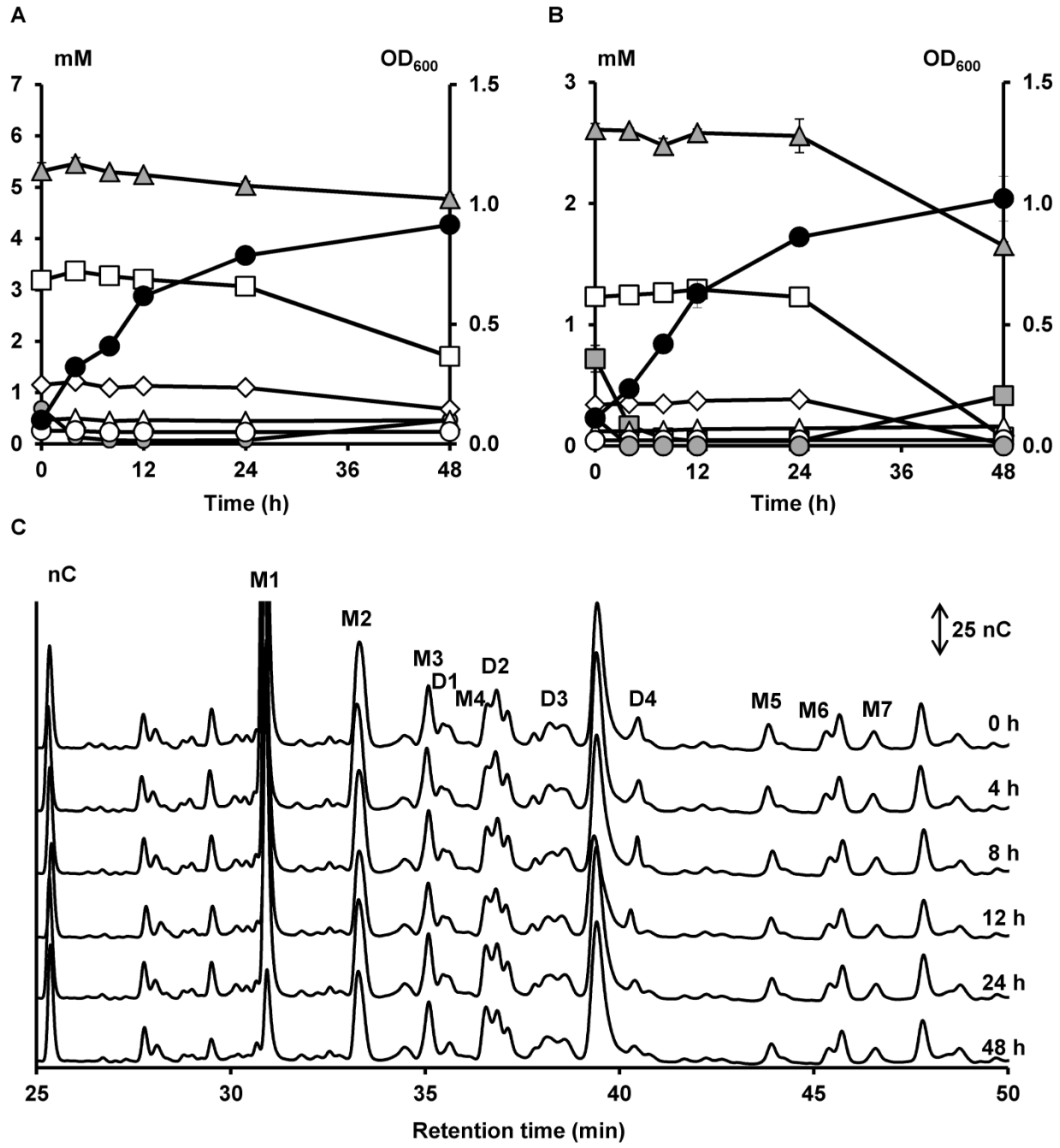

Figure S3.F. Growth of and carbohydrate degradation by *Bifidobacterium animalis* subsp. *lactis* BB-12 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

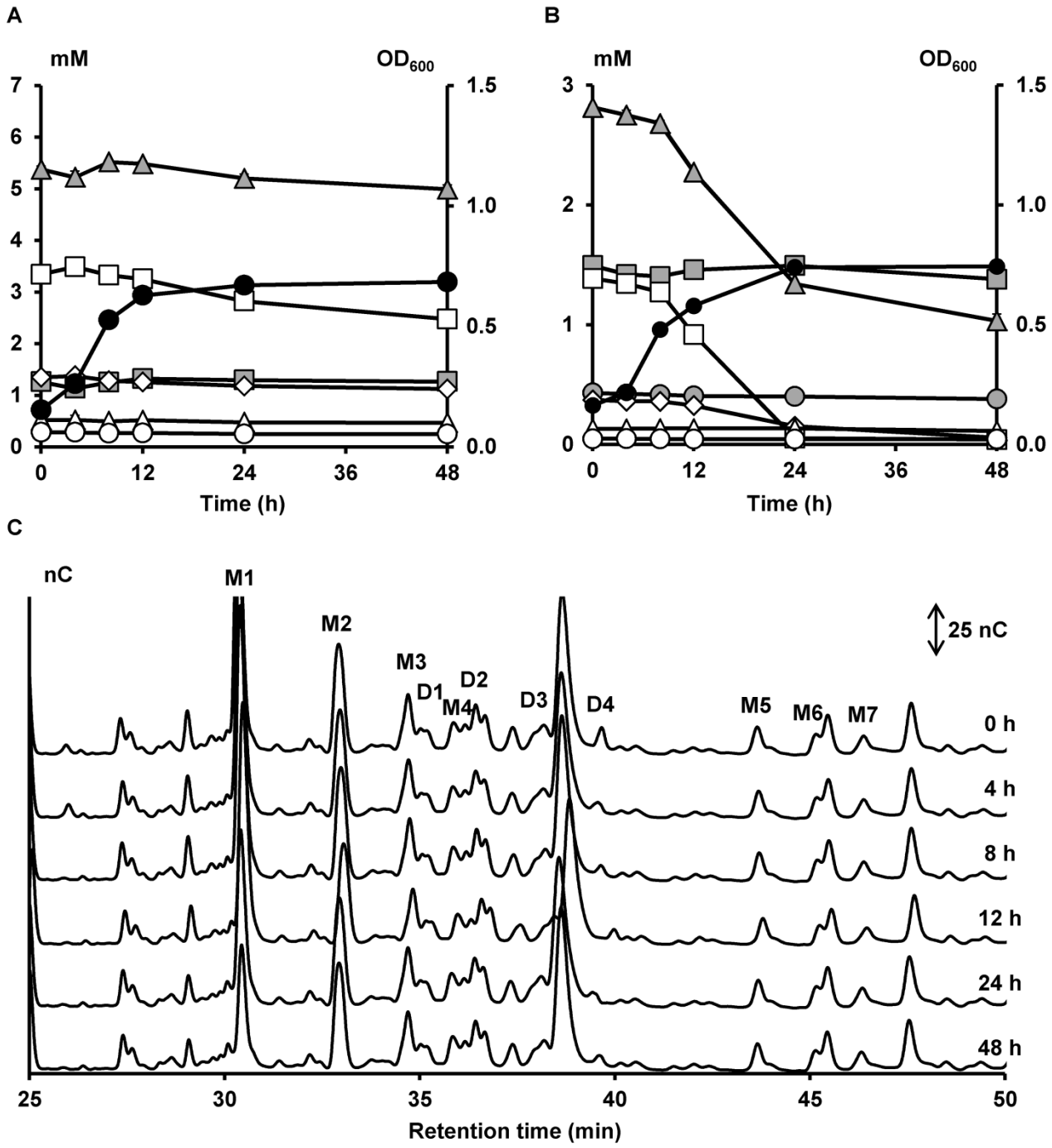

Figure S3.G. Growth of and carbohydrate degradation by *Bifidobacterium gallicum* LMG 11596^T in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

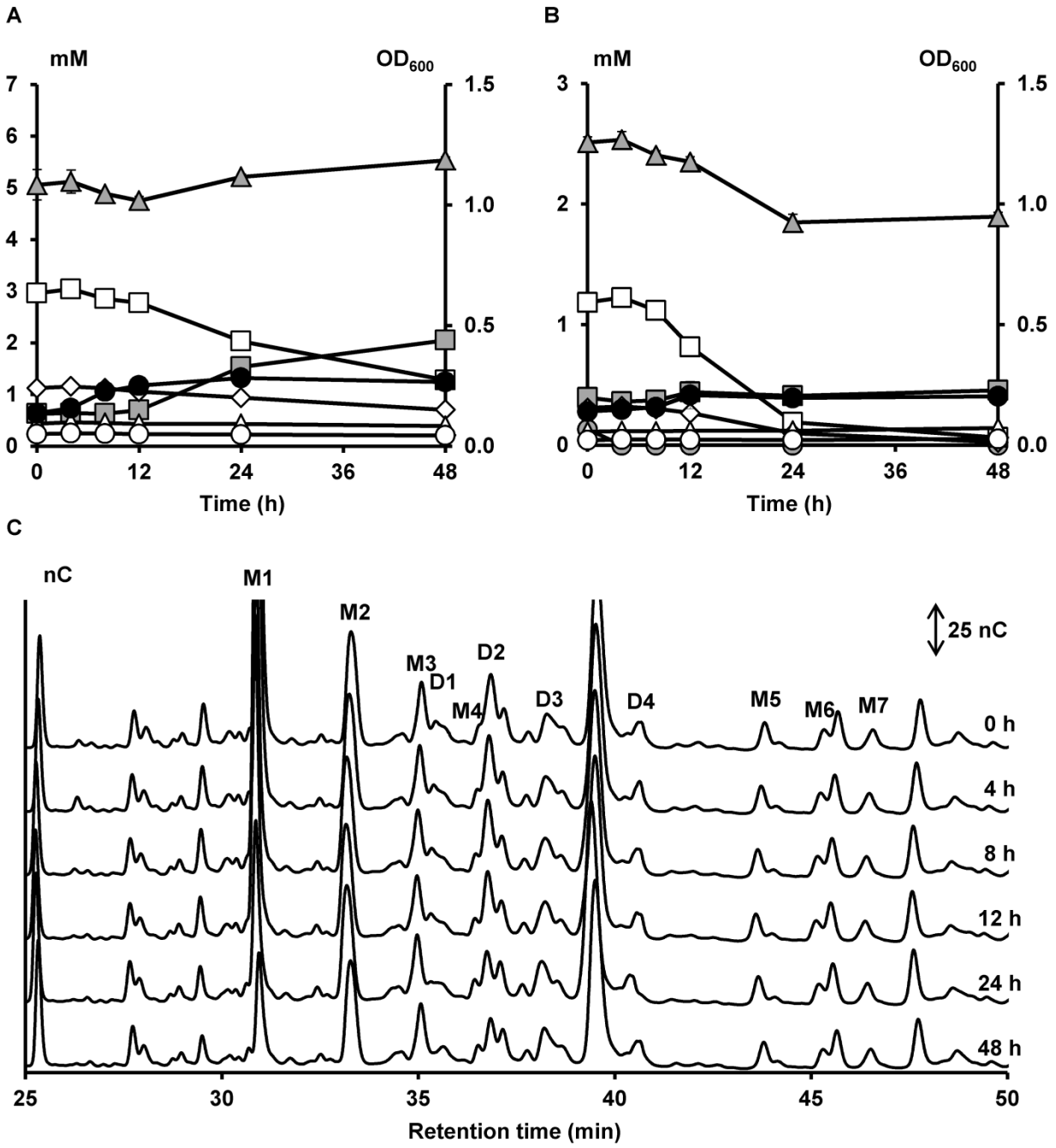

Figure S3.H. Growth of and carbohydrate degradation by *Bifidobacterium angulatum* LMG 11039^{T} in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

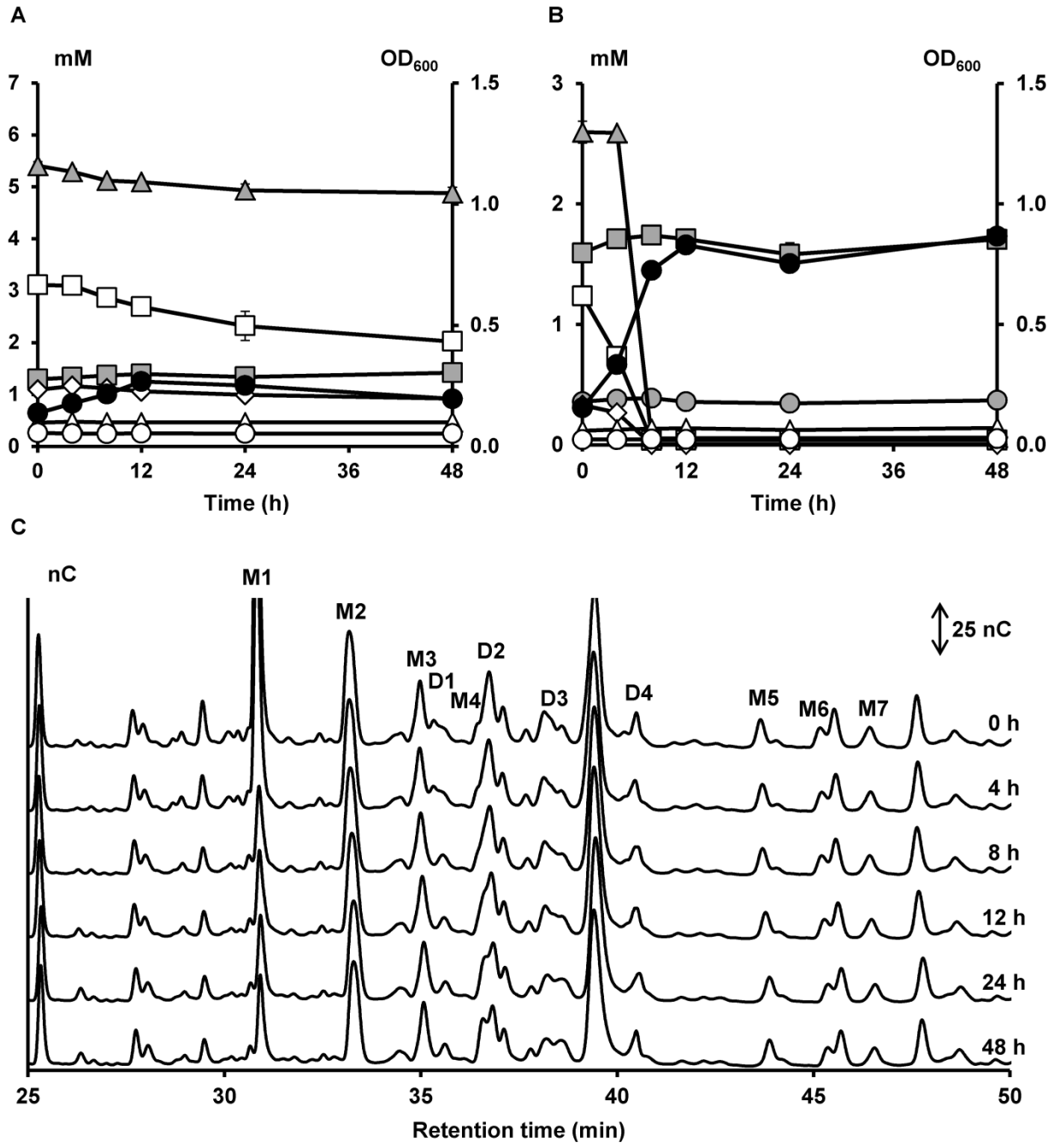

Figure S3.I. Growth of and carbohydrate degradation by *Bifidobacterium adolescentis* LMG 10502^T in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Peaks and symbols are named as in Figure S3.A.

Figure S4. Carbohydrate degradation by bifidobacterial strains belonging to cluster IV.

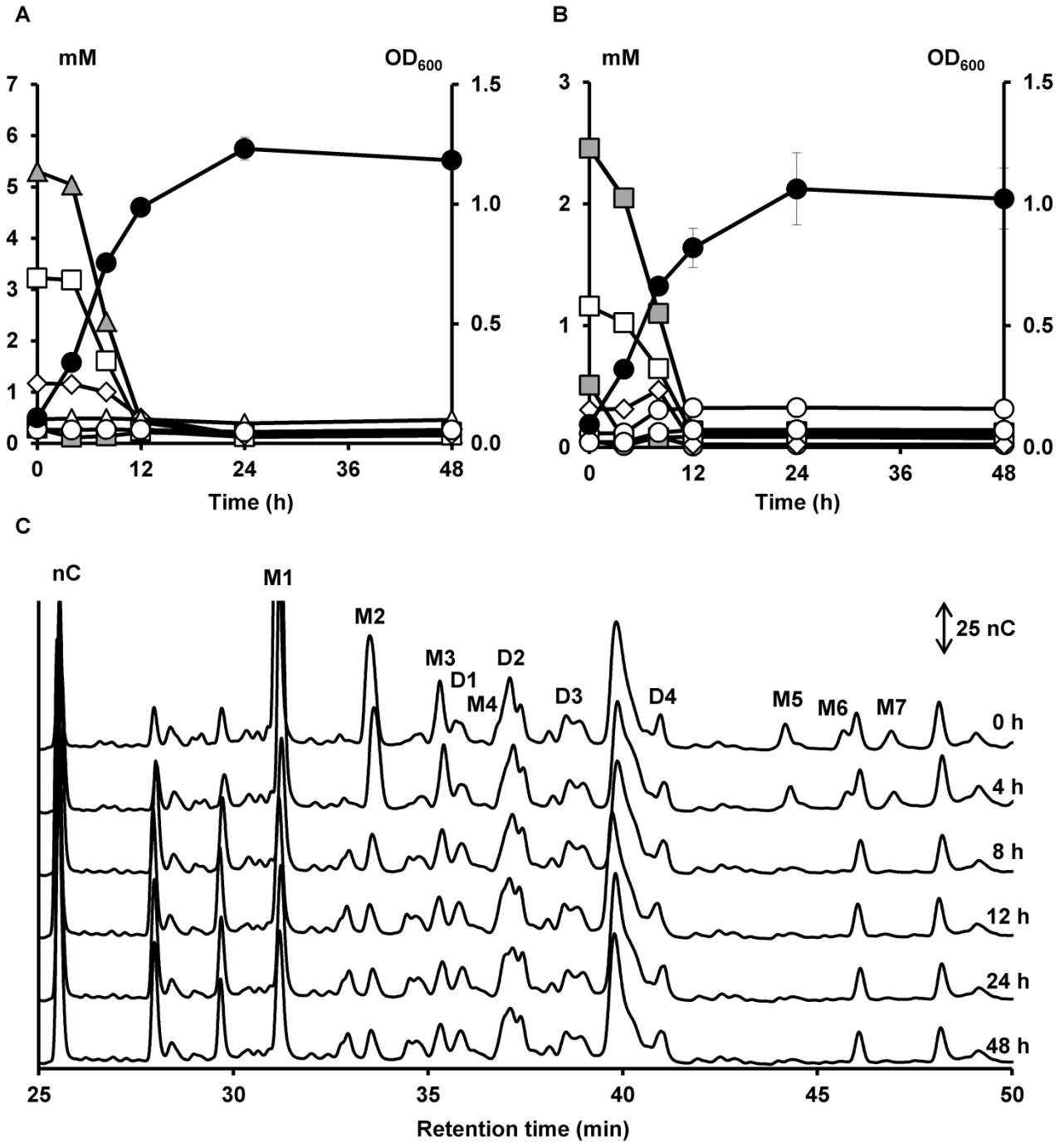

Figure S4.A. Growth of and carbohydrate degradation by *Bifidobacterium longum* subsp. *longum* CUETM 193 in a medium for colon bacteria with 5 g L^{-1} of XOS (A) or 5 g L^{-1} of (A)XOS (B and C) as a function of time (samples taken at 0, 4, 8, 12, 24, and 48 h). Growth is represented as OD_{600} measurements (●). Monosaccharide, XOS, and $XOS_{(A)XOS}$ consumption are represented as

residual concentrations of arabinose (●), xylose (■), xylobiose (▲), xylotriose (□), xylotetraose (◊), xylopentaose (Δ), and xylohexaose (○) in mM. (C) represents an enlargement of the AXOS HPAEC-PAD chromatograms between 25.0 and 50.0 min of elution. D1-D4, AXOS peaks containing at least one doubly substituted Xyl*p* residue. M1-M7, AXOS peaks containing at least one monosubstituted Xyl*p* residue.