

SUPPLEMENTARY MATERIALS

Table S1. Calibration equations used for monodisaccharide and fructan standards.

Standard	Slope	y-intercept	R ²	Range [†]
Long-chain fructan	1.2246	-0.7401	0.9984	0-25 µg/mL (2 µg/mL)
Short-chain fructan	2.8919	0.1402	0.9998	0-15 µg/mL (0.5 µg/mL)
Bifurcose	0.7964	0.0322	0.9995	0-2.5 µM (0.052 µM)
1-Nystose	1.9828	0.0166	0.9998	0-2 µM (0.042 µM)
6-Nystose	1.2009	-0.009	0.9994	0-2 µM (0.083 µM)
1-Kestose	1.8996	0.0075	0.9999	0-2 µM (0.042 µM)
6-Kestose	2.0009	0.0005	1	0-2 µM (0.042 µM)
Neokestose	1.5808	0.0163	0.9998	0-2 µM (0.042 µM)
Raffinose	2.025	0.0043	1	0-2 µM (0.042 µM)
Glucose	1.407	0.3399	0.9985	0-30 µM (0.21 µM)
Fructose	1.0131	0.512	0.995	0-30 µM (0.21 µM)
Sucrose	1.0867	0.2322	0.9967	0-15 µM (0.10 µM)

[†]Lowest concentration measured in calibration curve.

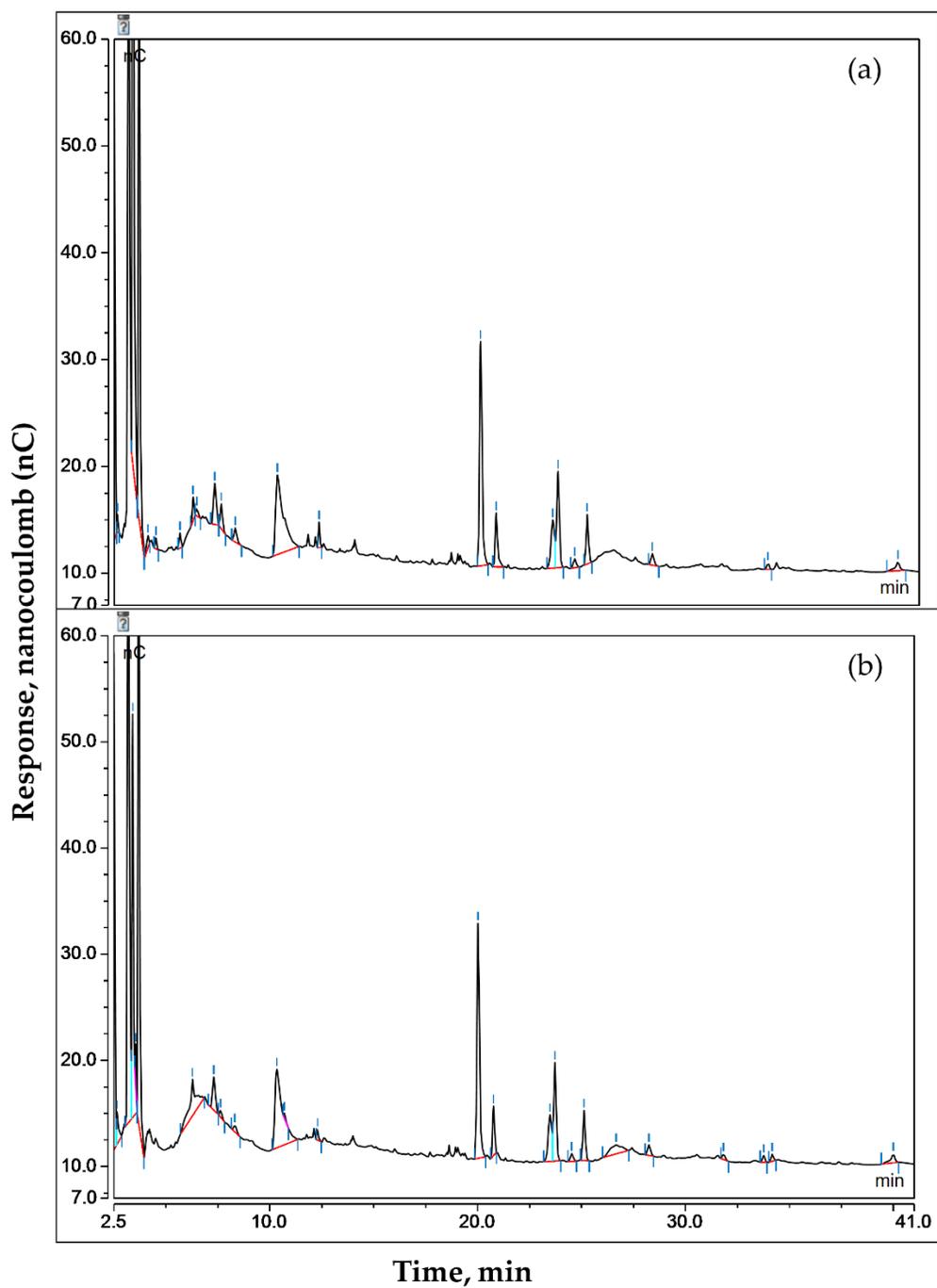


Figure S1. Representative chromatograms showing fructan profiles in rumen fluid-only controls following incubation with rumen fluid from one steer (a) or ram lamb (b). The areas of the isolated peaks present in controls were subtracted from the areas of peaks in the chromatograms of orchardgrass fermentations. Chromatograms were obtained by high-performance anion-exchange chromatography with pulsed amperometric detection (HPAEC-PAD).

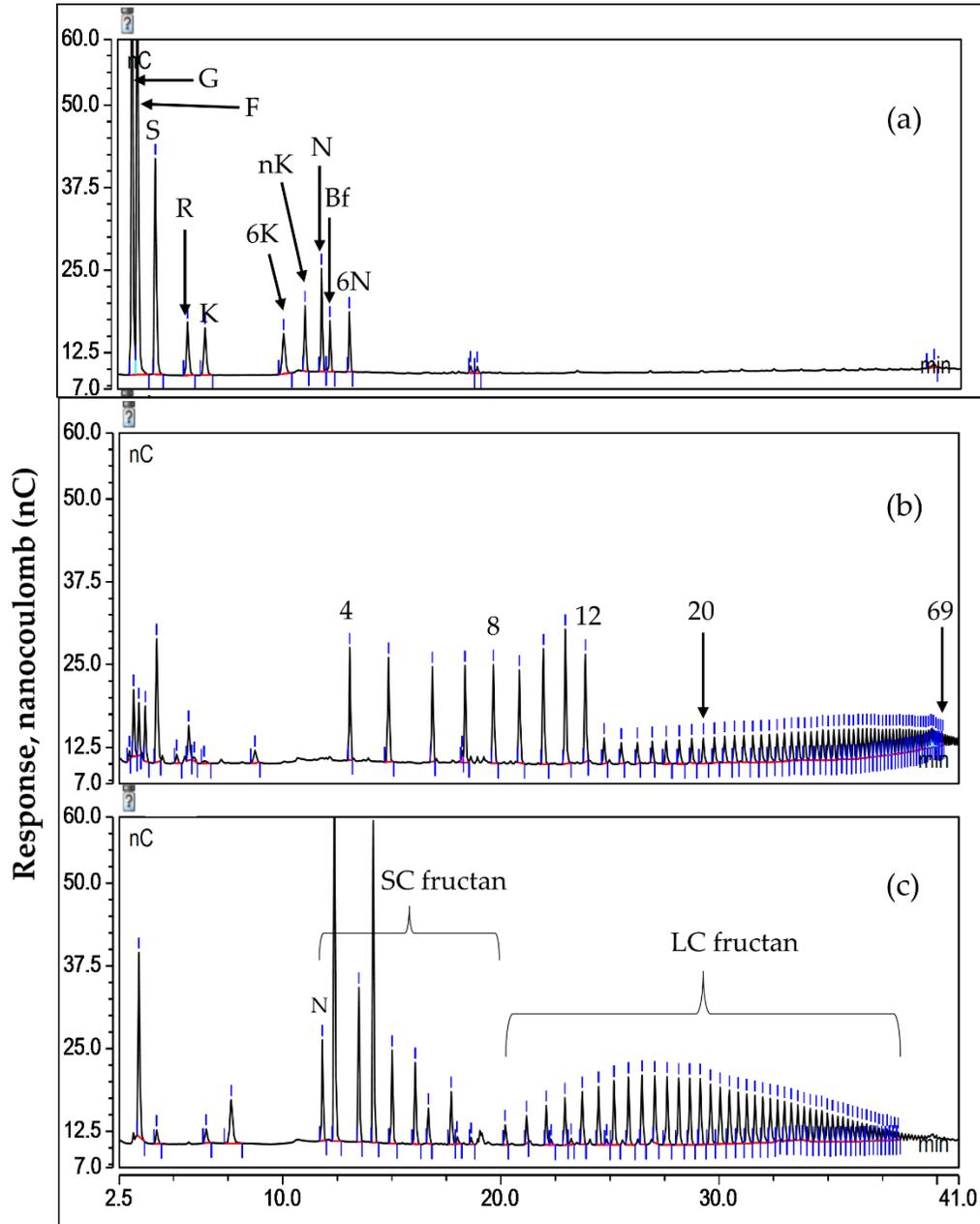


Figure S2. Chromatograms of the standards used in this study, including (a) a mixture of glucose (G), fructose (F), sucrose (S), trisaccharides raffinose (R), 1-kestose (K), 6-kestose (6K), neokestose (nK), 1-nystose or nystose (N), bifurcose (Bf), and 6-nystose (6N); (b) a mixture of fructans from big bluegrass (*Poa secunda* J. Presl), DP 4 to 12 (numbers above corresponding peaks), and fructans from orchardgrass (*Dactylis glomerata* L.) of DP ranging from about 7 to 69, used to determine DP in samples; and (c) a mixture of short-chain (SC) and long-chain (LC) fructan from chicory, used to quantify short- and long-chain fructans. The 1-nystose (N) peak is indicated by an arrow. Chromatograms were obtained by high-performance anion-exchange chromatography with pulsed amperometric detection (HPAEC-PAD).