

**Supplemental Table S2.** GC-MS/MS testing conditions for short chain fatty acids

(SCFAs) in rumen fluid sample

| Items                       | Parameter   |
|-----------------------------|---|
| Injection volume            | 2 $\mu\text{L}$   |
| Front Inlet Mode            | Splitless   |
| Carrier Gas                 | Helium  |
| Column                      | DB-FFAP (30 m $\times$ 0.25 mm $\times$ 0.25 $\mu\text{m}$ )  |
| Column Flow                 | 1.2 $\text{min}^{-1}$   |
| Oven Temperature Ramp       | Holds on 1 min at 95 $^{\circ}\text{C}$ , raises to 100 $^{\circ}\text{C}$ at a rate of 25 $^{\circ}\text{C}/\text{min}$ , raises to 130 $^{\circ}\text{C}$ at a rate of 17 $^{\circ}\text{C}/\text{min}$ , holds on 0.4 min, raises to 200 $^{\circ}\text{C}$ at a rate of 25 $^{\circ}\text{C}/\text{min}$ , holds on 0.5 min, running for 3 min. |
| Front Injection Temperature | 200 $^{\circ}\text{C}$  |
| Transfer Line Temperature   | 230 $^{\circ}\text{C}$  |
| Ion Source Temperature      | 230 $^{\circ}\text{C}$  |
| Quad Temperature            | 150 $^{\circ}\text{C}$  |
| Electron Energy             | 70 eV   |
| Scan mode                   | MRM   |
| Solvent Delay               | 3.0 min   |