

## COMPARISON OF THREE BUFFERS FOR THE SIMULATION OF SUBACUTE RUMINAL ACIDOSIS IN AN IN-VITRO RUMEN INCUBATION

### BACKGROUND

- Subacute ruminal acidosis (SARA) is characterized by a decreased ruminal pH for a longer period of the day.
- SARA is a well-recognized digestive disorder in most dairy herds, with a suggested incidence of 19% in early lactation dairy cows.
- An in-vitro simulation model offers opportunities to study some aspects of rumen metabolism while at the same time reducing the number of test animals.
- However, the buffering capacity of the standard phosphate-bicarbonate buffer used in-vitro is unable to prevent serious pH declines at a start pH below 6.0.

#### Collecting rumen fluid



#### Buffered rumen fluid



#### Injecting to anaerobic incubation flasks



#### Incubating at 39 °C



### GOAL

- To find a buffer which guarantees a stable pH throughout the in-vitro incubation period for both conditions with a pH range between 6.2 and 6.8 (normal condition) as well as in the pH range of 5.5 to 6.0 (simulation of a subacute ruminal acidosis).

### MATERIALS AND METHODS

- 50 mg dried maize silage was added to a 5 ml mixture of buffer : rumen fluid (4:1).
- Three biological buffers, 3.7 g/L tricarballylate buffer, 7.53 g/L Bis-tris buffer and 54.31 g/L 2-(N-morpholino) ethanesulfonic acid (MES) buffer were chosen based on their useful pH range.

#### Experiment 1

- The three buffers with pH adjusted to 6.8 and 5.8 separately were mixed with a rumen fluid mixture from three early lactating cows at a ratio of 4:1, incubated for 24 hours and 48 hours.

#### Experiment 2

- MES buffer at 6.8 and 5.8 pH were mixed separately with rumen fluid from either two early lactating or two dry cows. In vitro methane and hydrogen production, pH, Volatile fatty acid were measured at 0, 1.5, 3, 6, 24, and 48 hours after incubating.

### RESULTS

Fig.1. pH value during incubations at pH of 6,8 or 5,8 at the start of the incubation

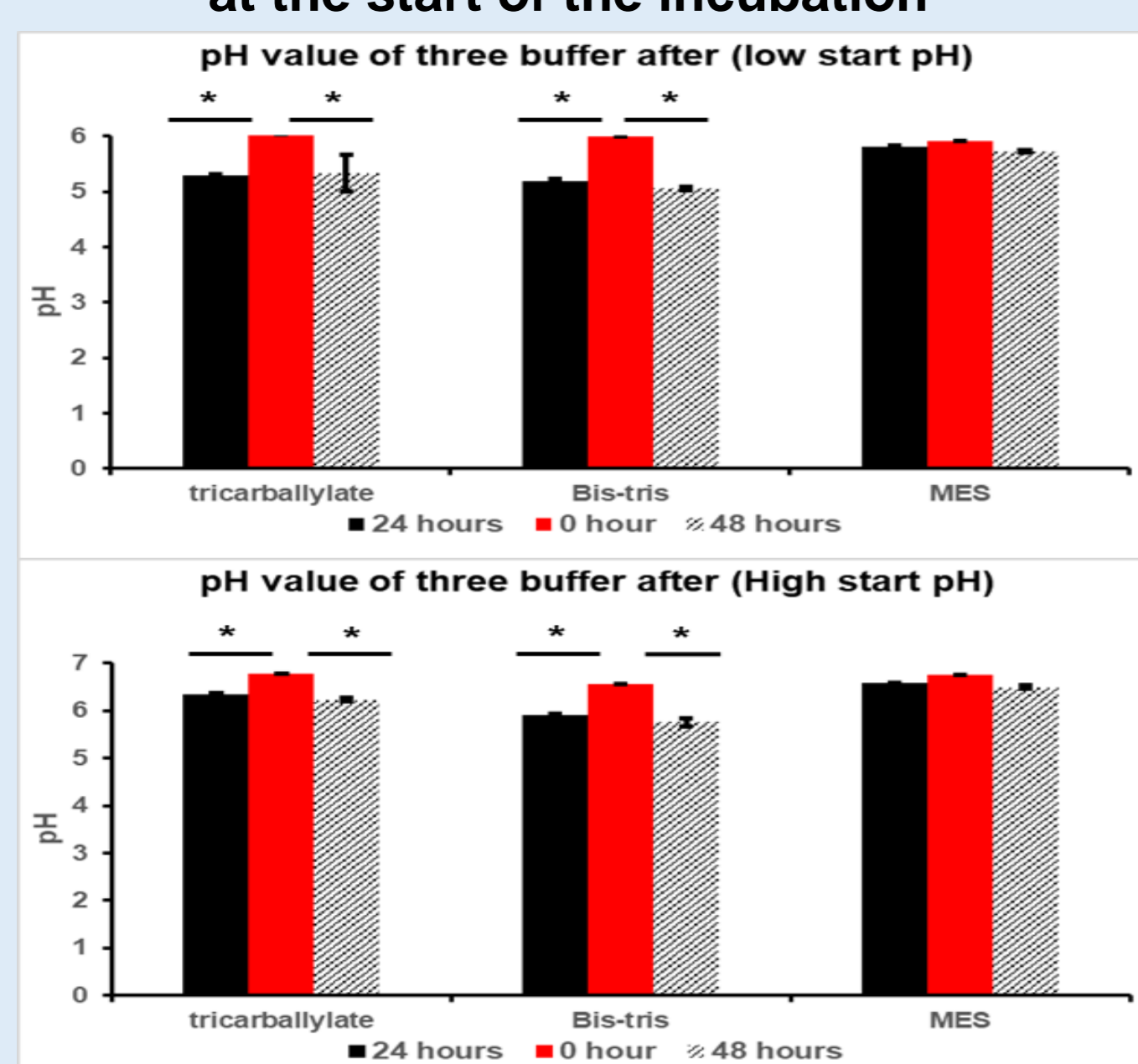
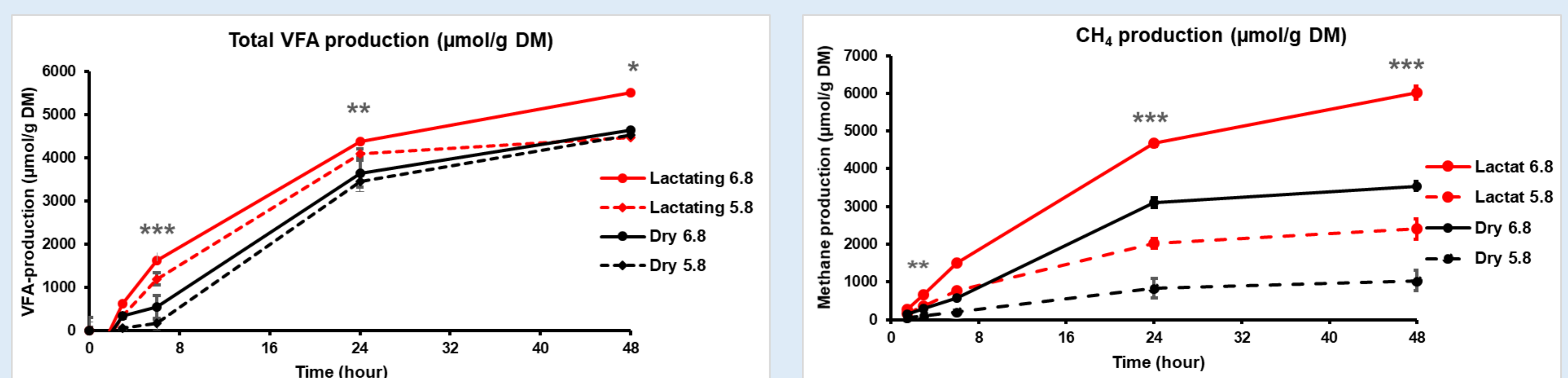


Fig.2. Total VFA or methane production ( $\mu\text{mol/g DM}$ ) during incubations with inoculum of either early lactating or dry cows in combination with a MES-buffer at pH of 6,8 or 5,8 at the start of the incubation



### CONCLUSION

- The pH in a MES buffer with start pH below 6.0 remained stable after 24 hours and 48 hours incubation.
- In the high pH simulation (start pH = 6.8), the MES buffer allowed to keep a stable pH after 24 and 48 hours of incubation.
- At each time point, the methane and VFA production of the high pH both with lactating and dry cows' inoculum was greater than in the low pH incubation.
- The MES buffer allows to simulate the pH range from normal to SARA conditions when incubated between 1.5 and 48 hours.

#### Contact

Hong. Yang@ugent.be  
www.lanupro.ugent.be

Universiteit Gent

@ugent

Ghent University