

## The Impact of Yeast Assimilable Nitrogen Concentration and Composition on Fermentation Kinetics and Hydrogen Sulfide Production during Cider Fermentation

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### Abstract

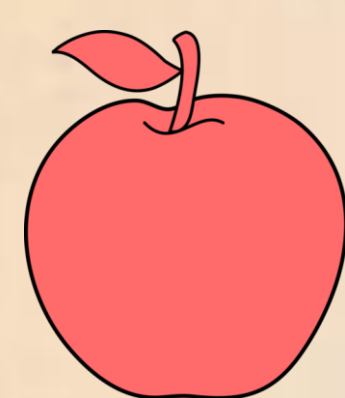
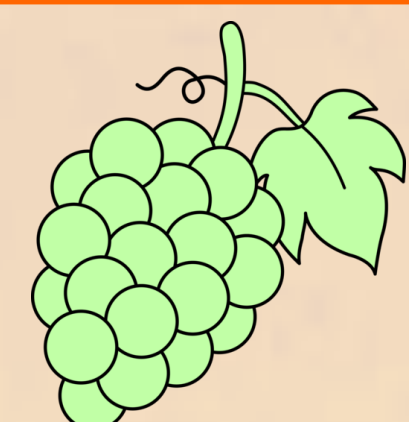
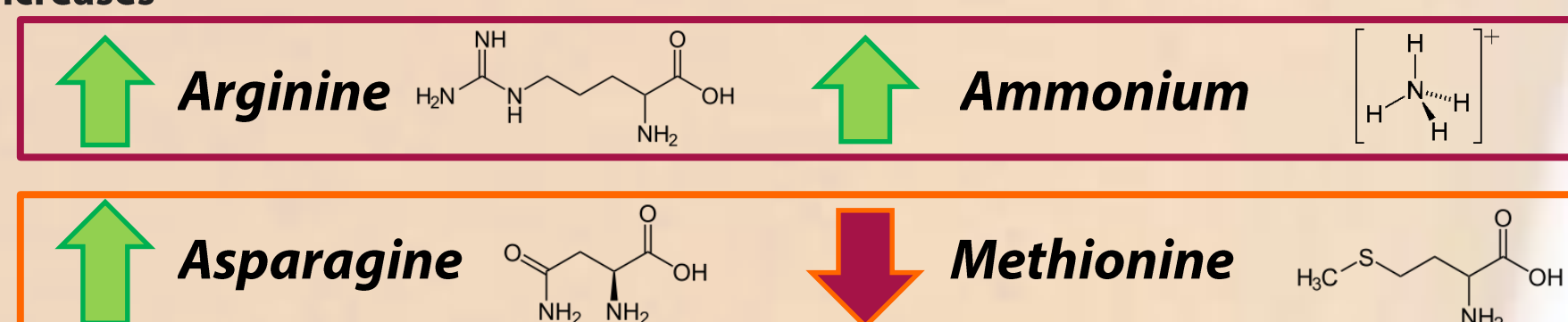
Yeast assimilable nitrogen (YAN) concentration and composition is known to significantly impact hydrogen sulfide (H<sub>2</sub>S) production and fermentation kinetics during wine fermentation but has not been extensively studied in cider fermentations. Nitrogen-deficient apple juice was supplemented with asparagine, arginine, methionine and ammonium and fermented with two yeast strains. There was no difference in H<sub>2</sub>S production between sources of nitrogen added as asparagine, arginine, and ammonium. Additions of methionine decreased H<sub>2</sub>S production at concentrations as low as 5 mg L<sup>-1</sup> in nitrogen-deficient juice in one yeast strain, but only high concentrations of methionine decreased H<sub>2</sub>S production when juice was supplemented with moderate YAN concentrations, and did not influence H<sub>2</sub>S production at high YAN concentrations. When juice was supplemented to 153 mg L<sup>-1</sup> YAN there was an increase in H<sub>2</sub>S production regardless of methionine treatment. H<sub>2</sub>S production decreased when juice was supplemented to 253 mg L<sup>-1</sup> YAN. Following sensory evaluation, panelists were able to discern differences in the aroma of samples supplemented with ammonium and methionine, which corresponds to an increase and decrease in H<sub>2</sub>S production during fermentation, respectively.

### Background & Objectives

**Hydrogen sulfide (H<sub>2</sub>S)** is a prominent off-aroma formed during cider fermentations as compared to wine fermentations, suggesting differences in apple and grape chemistry may lead to differential H<sub>2</sub>S production. One possible cause of H<sub>2</sub>S is **yeast assimilable nitrogen (YAN)** concentration and composition. The most prominent YAN components in grapes are typically **arginine** and **ammonium**, where as the most prominent YAN component in apples is **asparagine**. Additionally, grapes typically contain higher concentrations of **methionine**, which is known to decrease H<sub>2</sub>S, as compared to apples. As a whole, low YAN concentrations are known to increase H<sub>2</sub>S production. The objective of this study is to investigate these differences in apple and grape chemistry on H<sub>2</sub>S production in ciders.

#### Hypotheses

- Additions of **asparagine** will lead to **higher H<sub>2</sub>S production** as compared to additions of arginine or ammonium
- Additions of **methionine** will lead to **lower H<sub>2</sub>S production**
- Additions of **methionine will decrease H<sub>2</sub>S production** when total **YAN concentration increases**



### Results

Figure 1 – Total H<sub>2</sub>S production during fermentations with added methionine\*

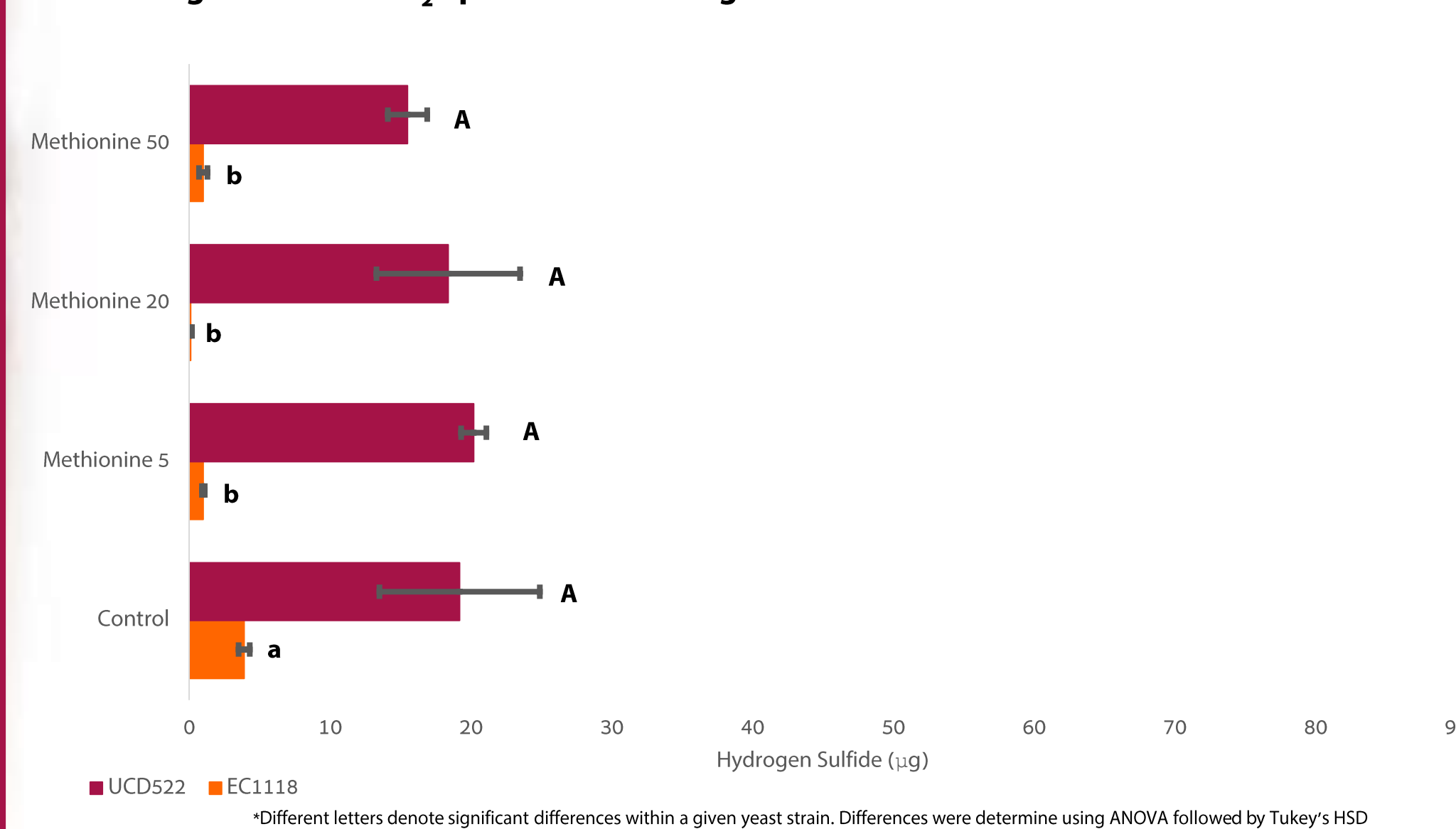
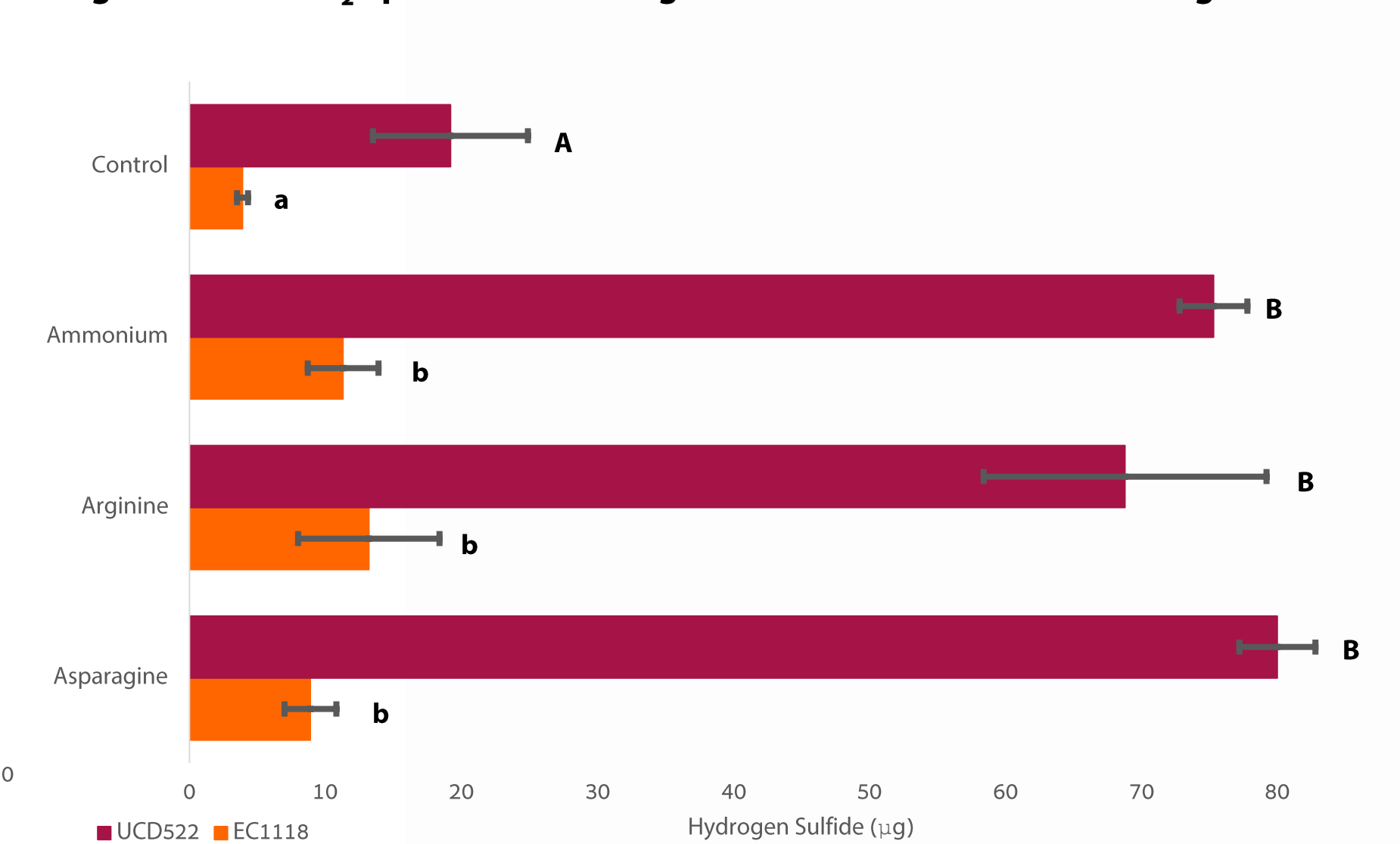


Figure 2 – Total H<sub>2</sub>S production during fermentations with added nitrogen sources\*



### Methods

Laboratory-scale fermentations were conducted with commercially-produced apple juice fermented in 200 mL aliquots. Two yeast strains were used to represent low (EC1118) and high (UCDS22) H<sub>2</sub>S-producing commercial yeasts. Fermentation treatments were conducted in triplicate. Fermentation mass was measured twice daily to monitor fermentation rate as a proxy for CO<sub>2</sub> production. H<sub>2</sub>S production was quantified by affixing a silica-packed aspirator tube as an airlock to the fermenter. CO<sub>2</sub> formed during fermentation carried H<sub>2</sub>S through the silica, creating a darkened band proportional to the total mass of H<sub>2</sub>S produced.

Fermentations were conducted in the following treatments in two yeast strains each:

- Control with no added nitrogen
- 140 mg N/L each asparagine, arginine and ammonium
- 5, 20 and 50 mg/L methionine

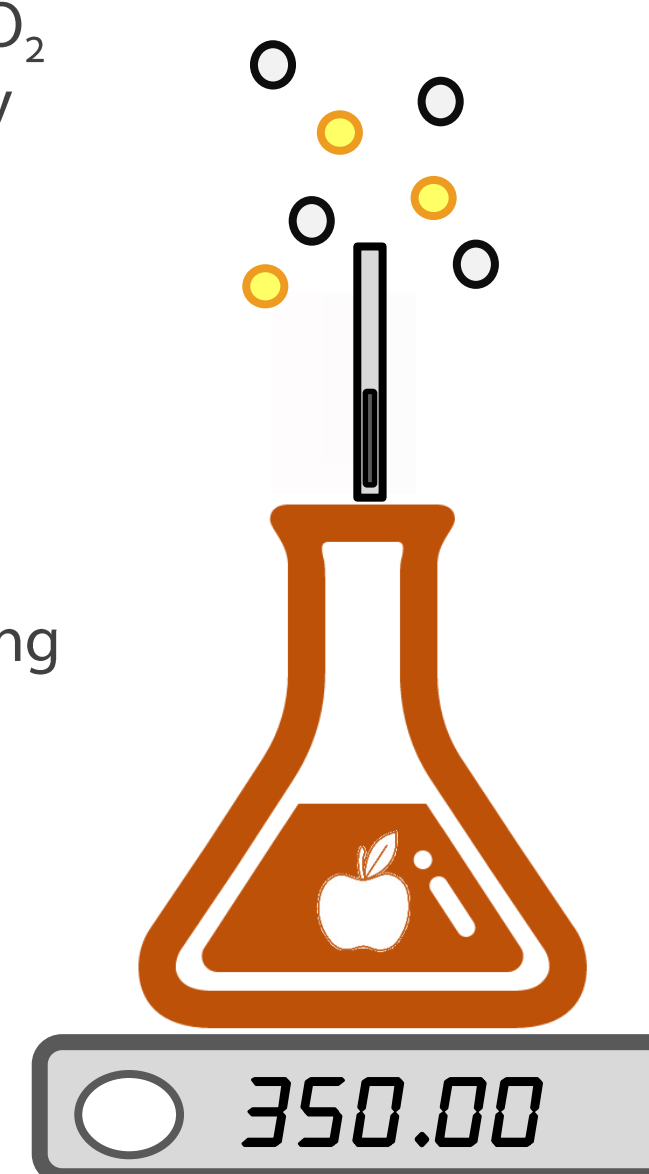


Figure 3 – Total H<sub>2</sub>S production during fermentations with added methionine at 3 YAN concentrations\*

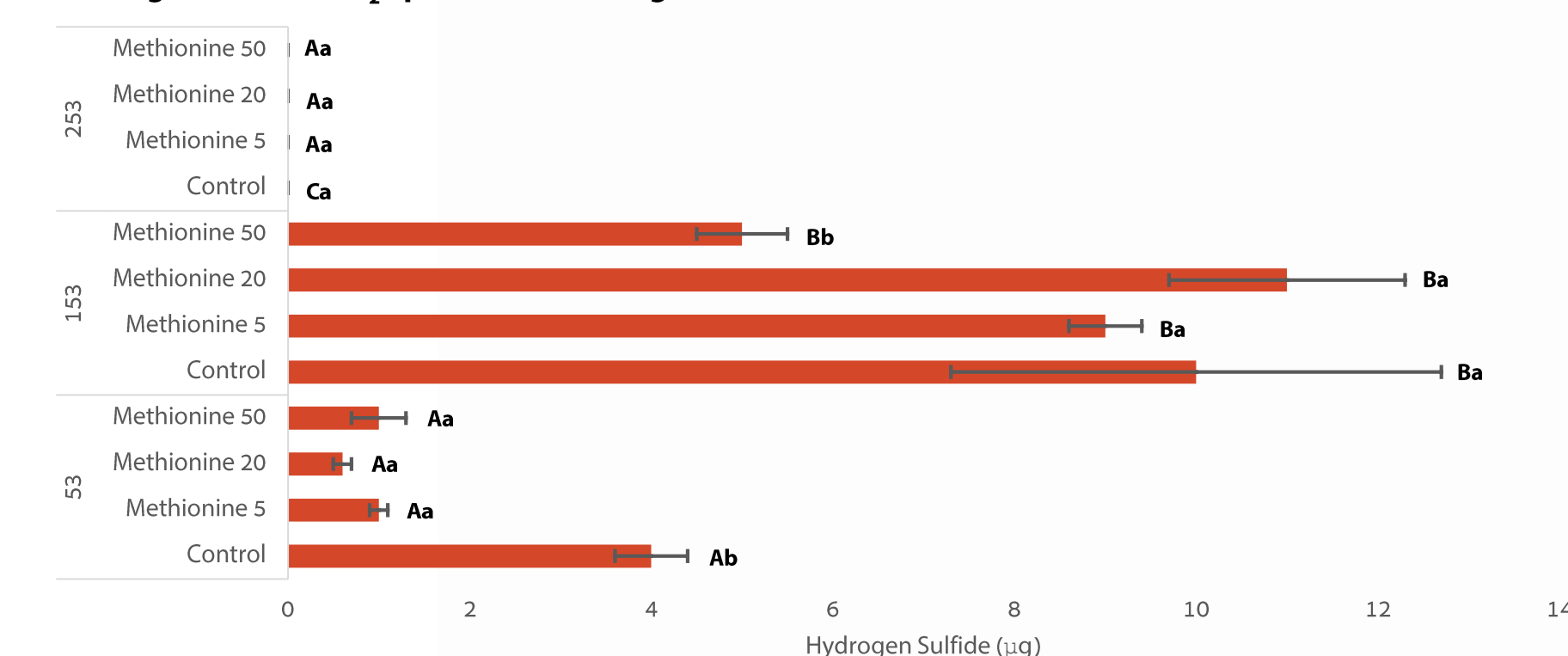
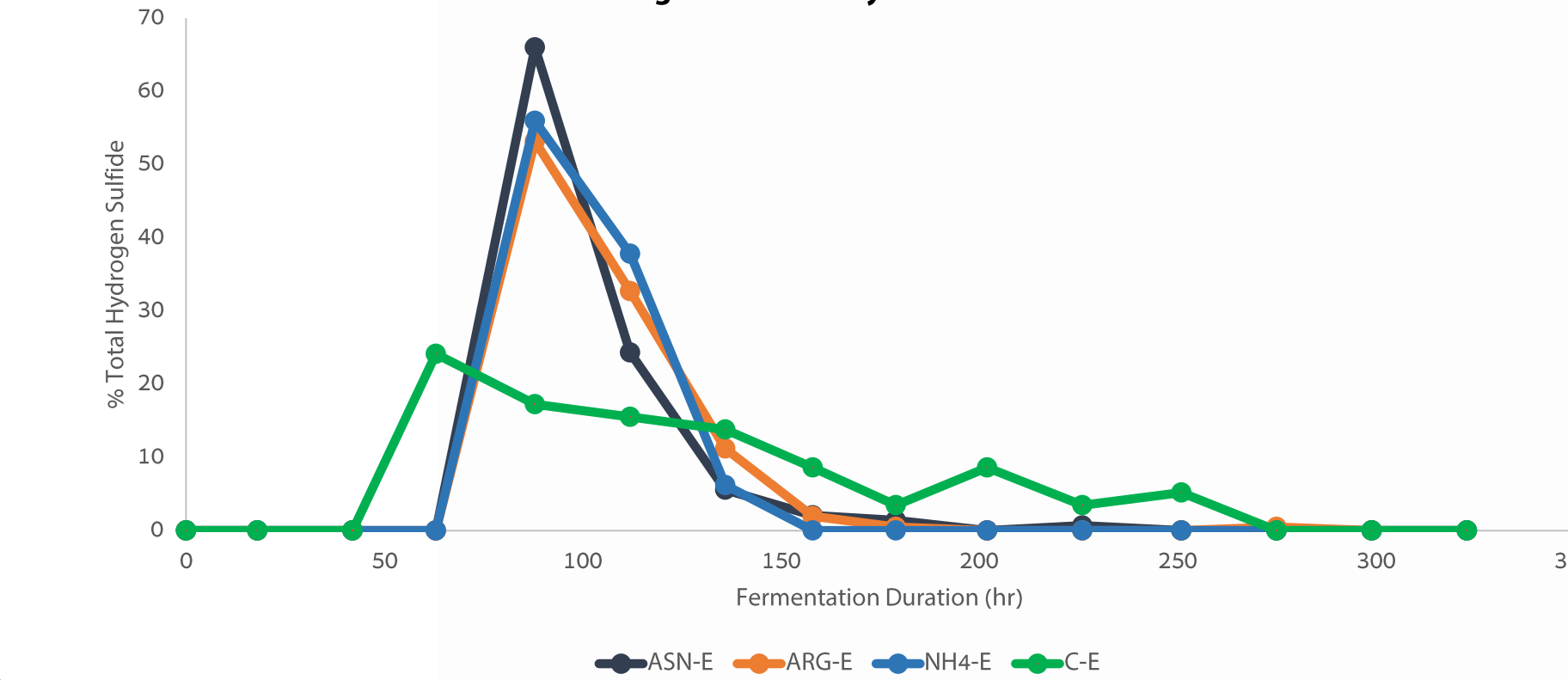
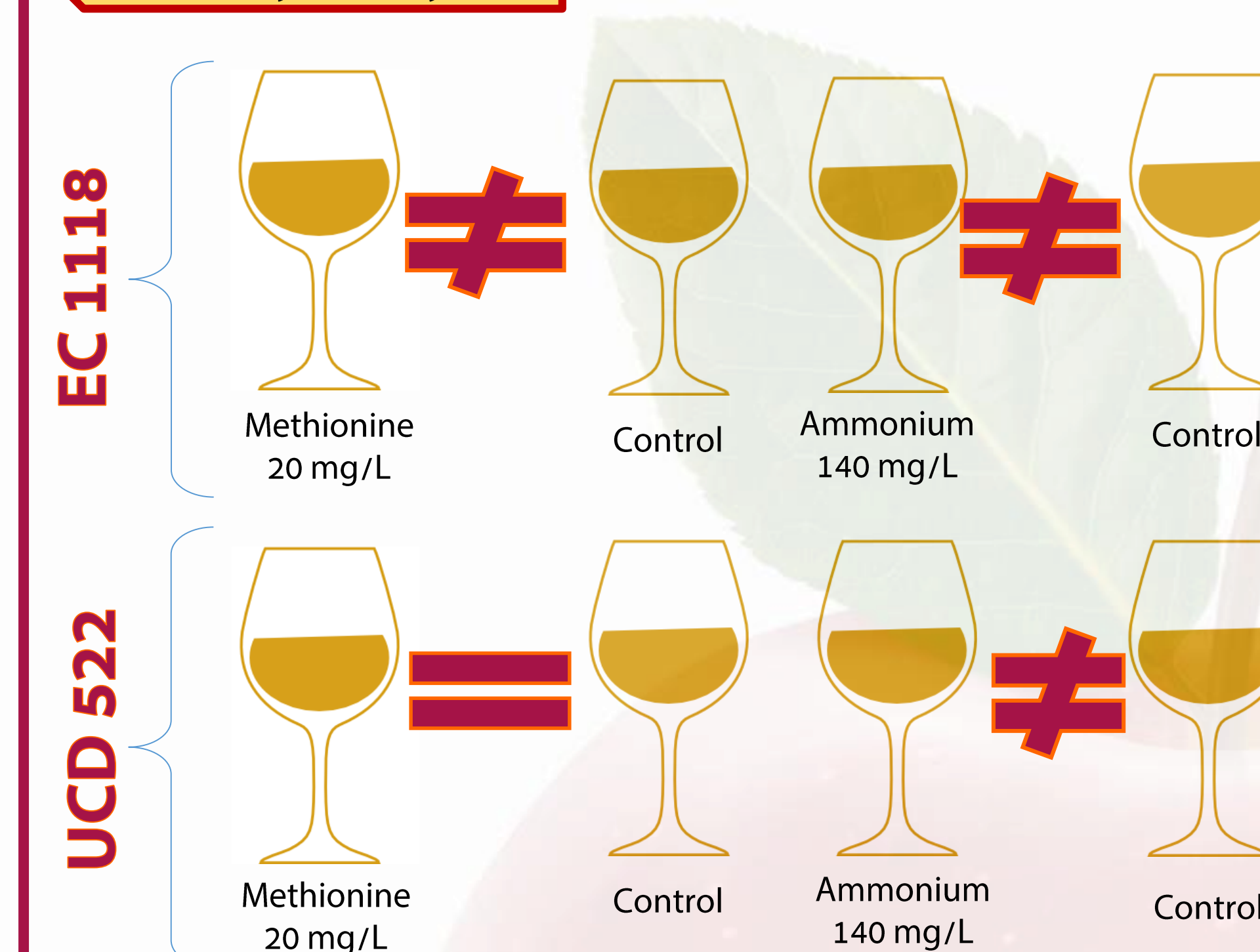


Figure 4 – Percent H<sub>2</sub>S produced during the time course of fermentation for fermentations with added nitrogen sources in yeast strain EC1118



### Sensory Analysis



Aroma difference testing (triangle test) was conducted on 4 sample sets, with the following results:

- EC1118**
  - Methionine at 20 mg/L (0.6 µg H<sub>2</sub>S) ≠ Control (4 µg H<sub>2</sub>S)
  - NH<sub>4</sub> at 140 mg N/L (11 µg H<sub>2</sub>S) ≠ Control (4 µg H<sub>2</sub>S)
- UCD522**
  - Methionine at 20 mg/L (19 µg H<sub>2</sub>S) = Control (18 µg H<sub>2</sub>S)
  - NH<sub>4</sub> at 140 mg N/L (75 µg H<sub>2</sub>S) ≠ Control (18 µg H<sub>2</sub>S)

### Conclusion

Based on the results of this study and given our hypotheses, we can make the following conclusions:

- There is **no difference in H<sub>2</sub>S production** between nitrogen sources **ammonium, arginine and asparagine**
- In one yeast strain, additions of **methionine decrease H<sub>2</sub>S** at all concentrations
- Increasing YAN concentration decreases the beneficial effects of methionine**
- Contrary to general practice, **moderate YAN concentrations (153 mg/L) increased H<sub>2</sub>S** as compared to low (53 mg/L) or high (253 mg/L) YAN
- There are **perceived aroma differences** correlating to treatments with both **increased and decreased H<sub>2</sub>S production**

These results indicate that the differences in apple and grape chemistry may lead to differences in H<sub>2</sub>S production during cider fermentation. Although primary nitrogen source does not affect H<sub>2</sub>S production, low concentrations of methionine observed in apples may lead to increased H<sub>2</sub>S production. Cidermakers should be aware of the adverse effects of low YAN concentrations on cider aroma and quality. The beneficial effects of methionine may lead to the production of yeast nutrients which can inhibit H<sub>2</sub>S production during cider fermentation.

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