69th EAAP Annual Meeting

Variation in the solubilization of nitrogenous compounds in wheat straw by different white-rot fungi

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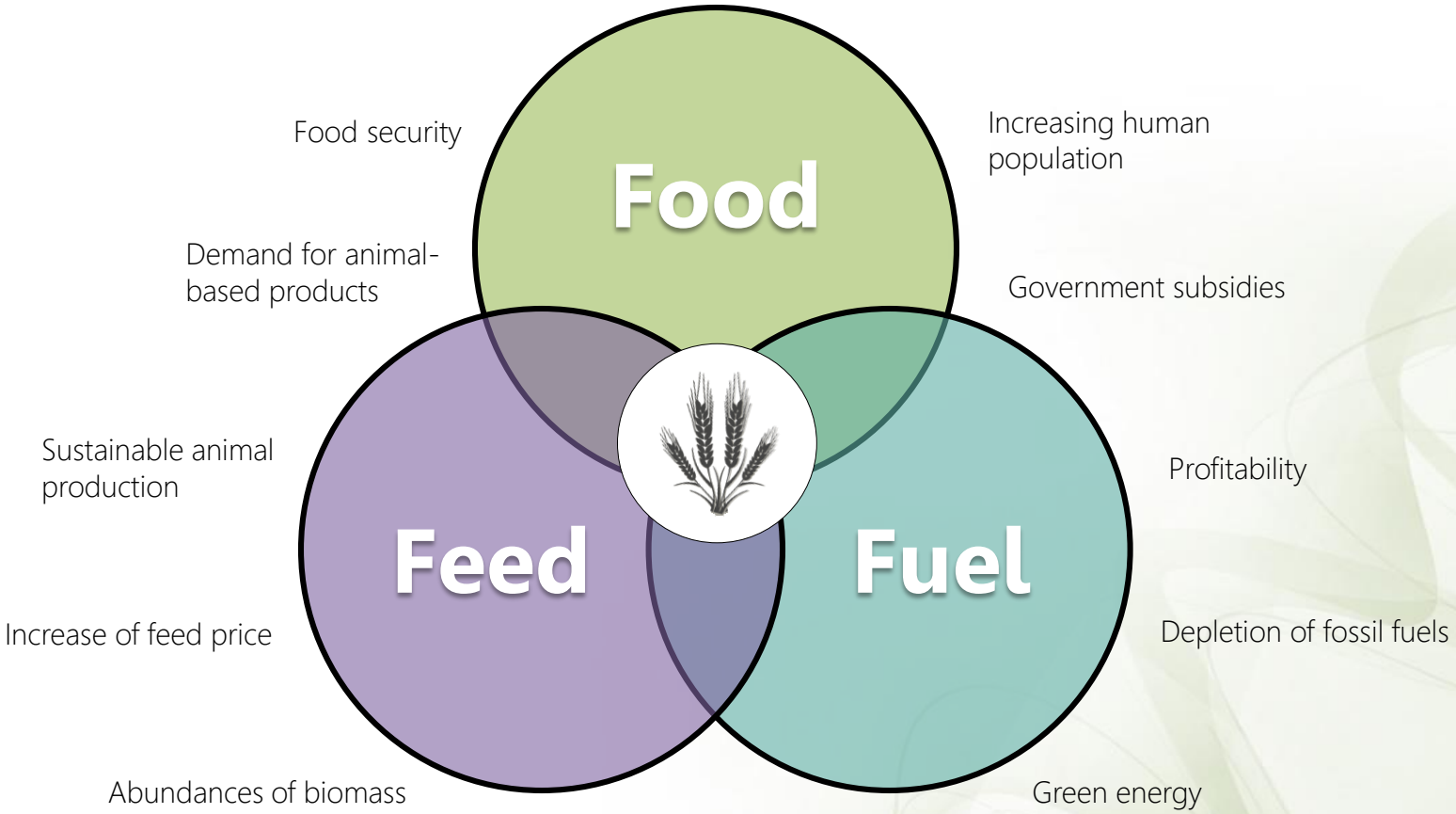
Background

Sustainability issues



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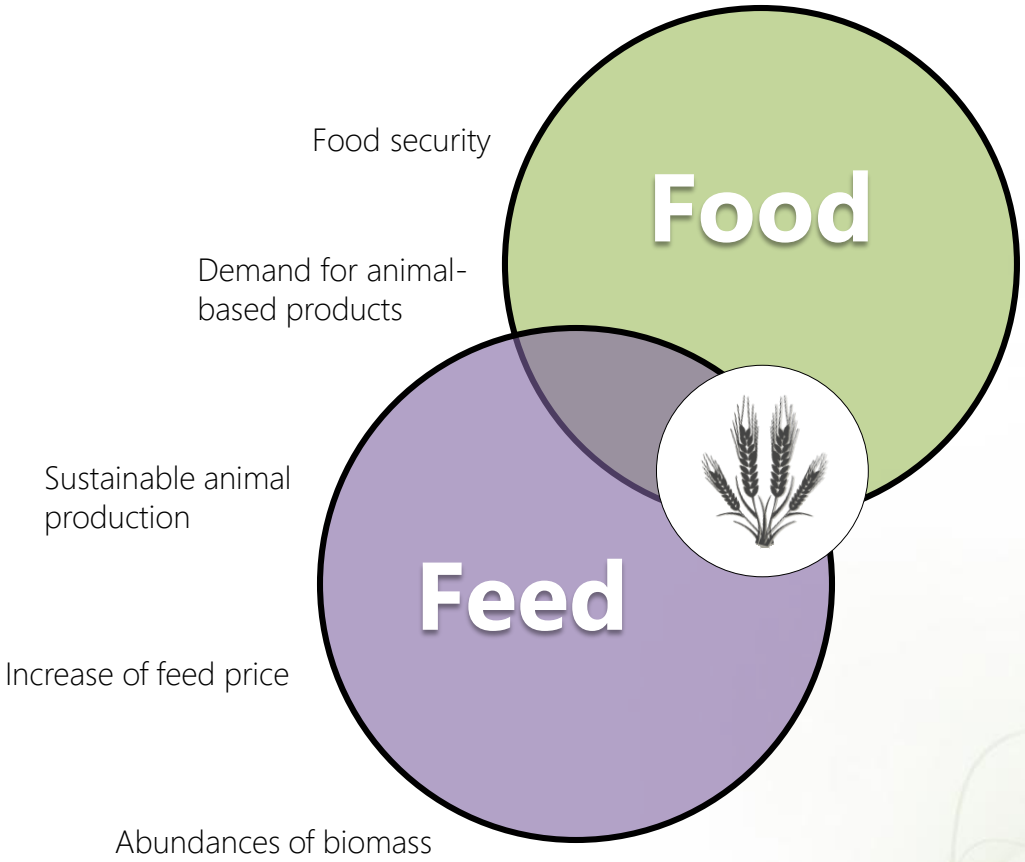
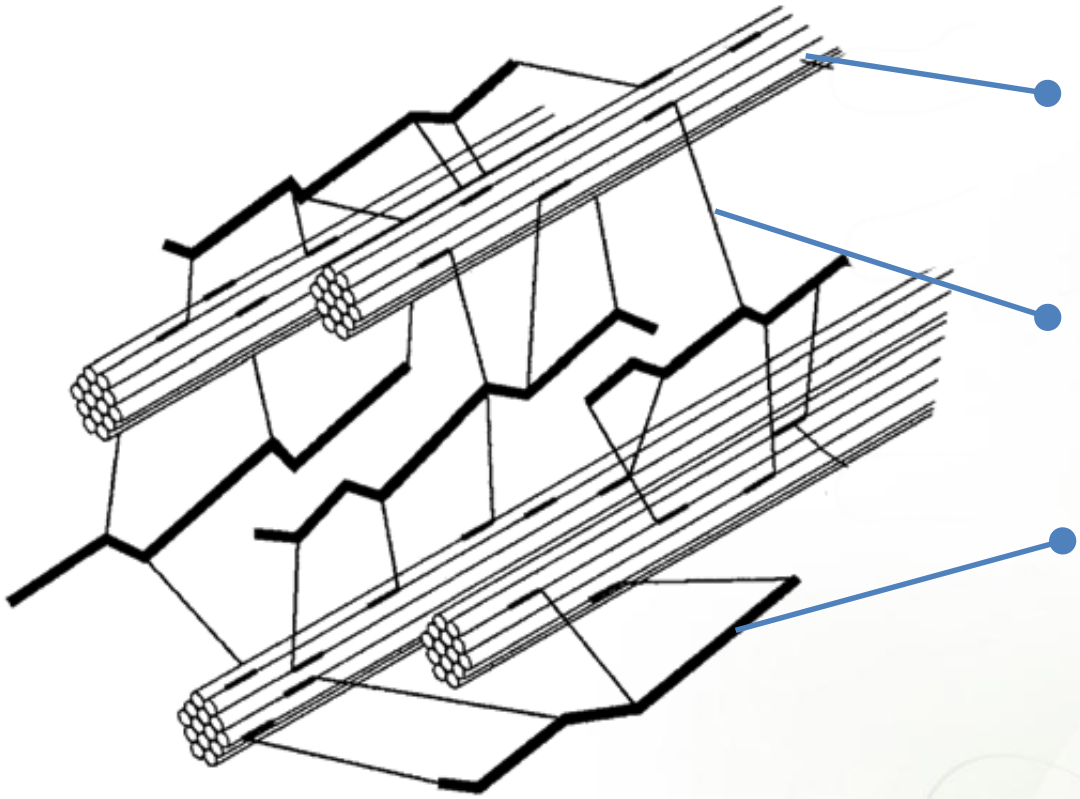


Background

Unlocking the lignin-carbohydrates complex



Cellulose
46.1 to 50.0%

Hemicellulose
26.0 to 33.8%

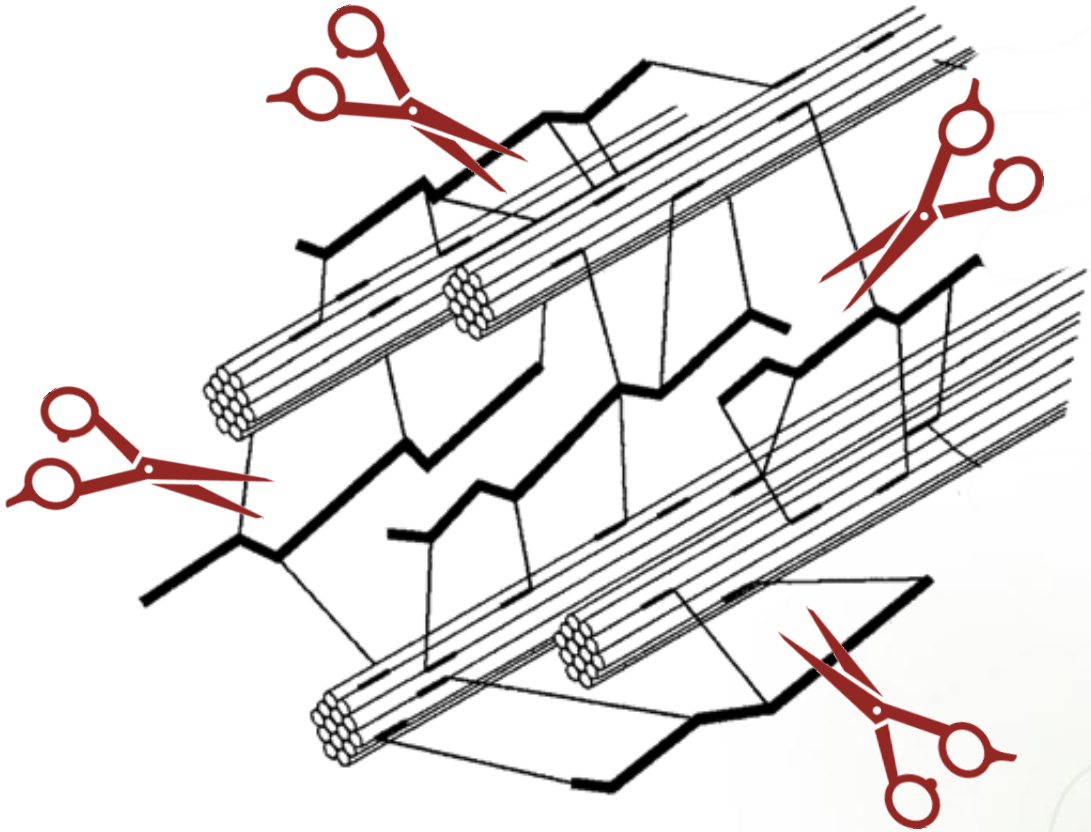
Lignin *
5.6 to 9.2%

Protein
1.1 to 3.3%

* Acid detergent lignin; Data obtained from 3 independent experiments

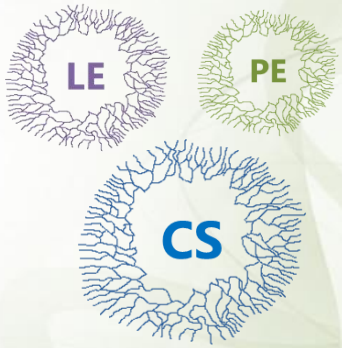
Background

Unlocking the lignin-carbohydrates complex



Physical + **Chemical**

Biological



Hendriks et al., **Bioresour.** (2009); Hatakka & Hammel, **The Mycota** (2010)

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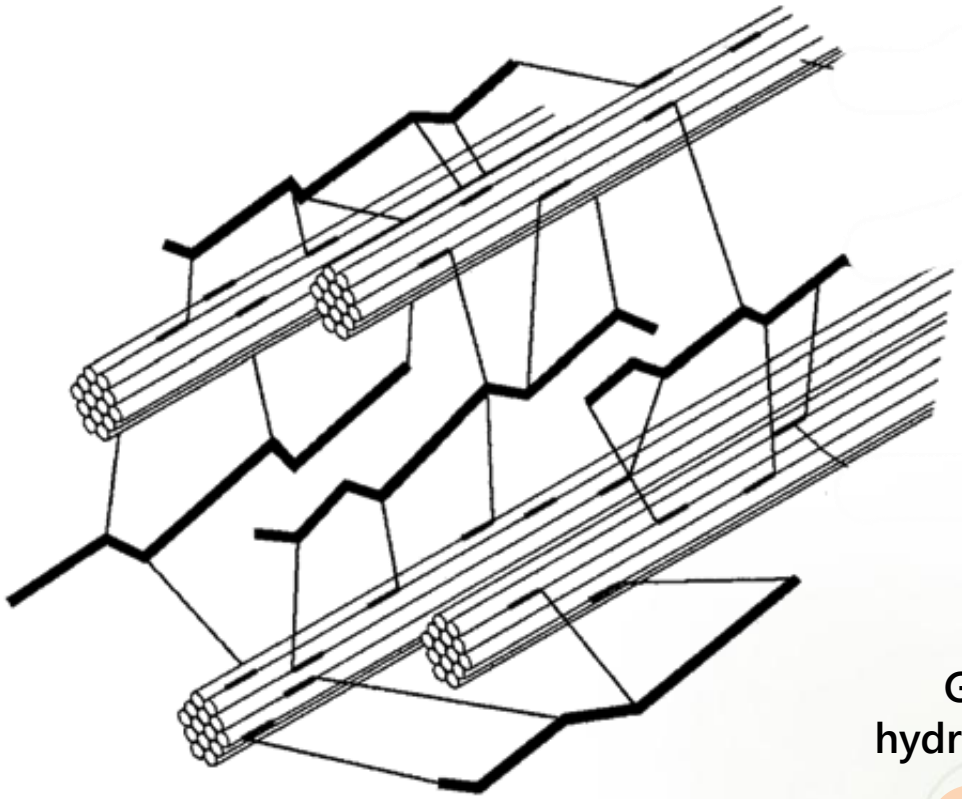

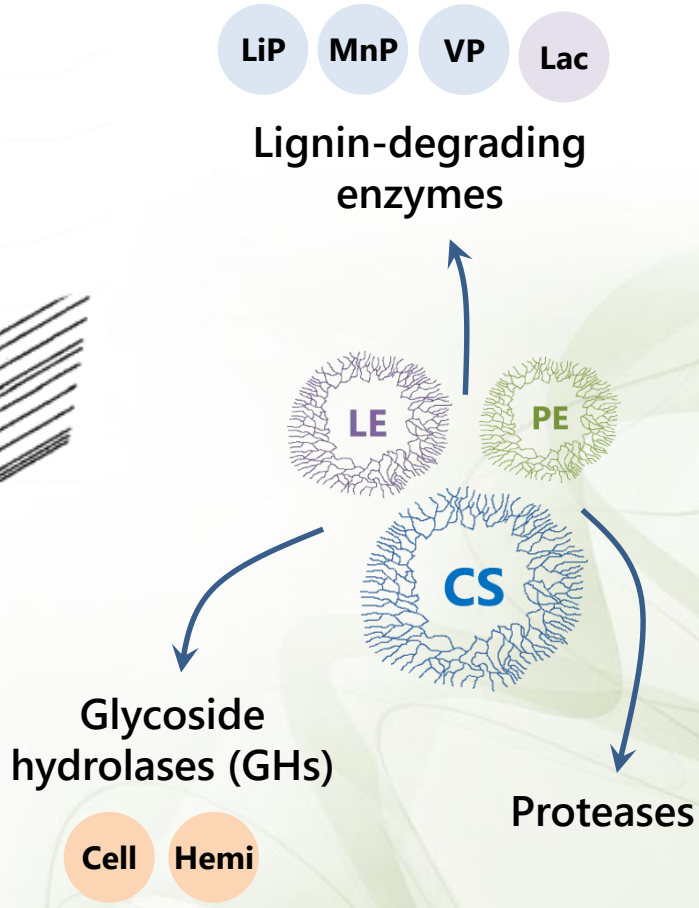

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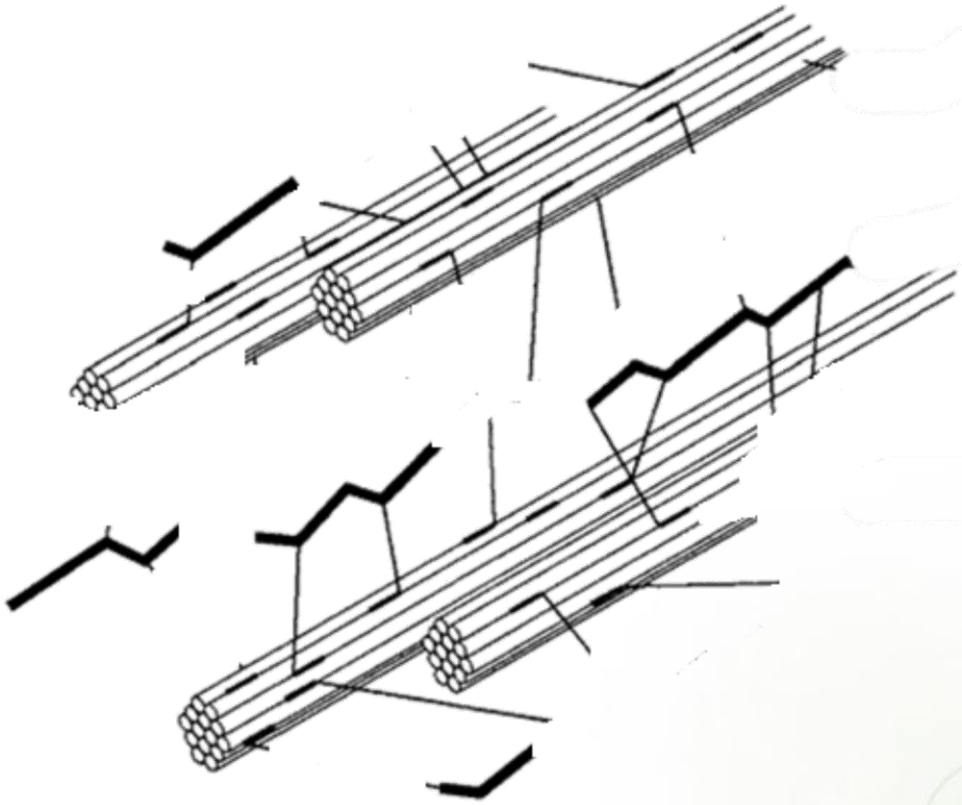


↓ Lignin

↑ Available carbohydrates

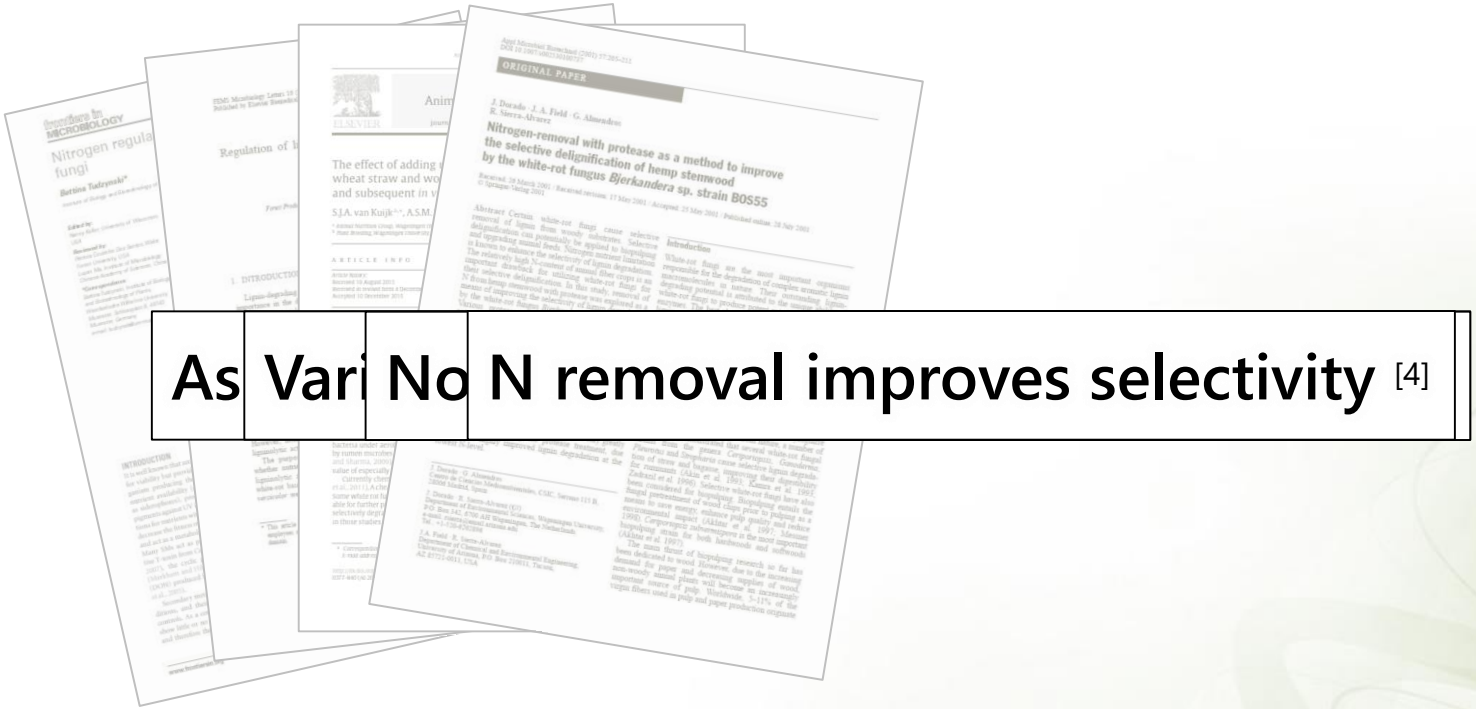
↑ *In vitro* Degradability

? Protein availability

Tuyen et al., Bioresour. (2012); van Kuijk et al., J. Anim. Sci. Biotechnol (2016)

Background

Nitrogen metabolism by fungi



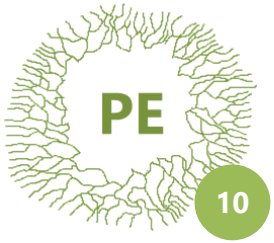
[1] Tudzynski, **Front. Microbiol**. (2014); [2] Leatham & Kent , **FEMS Microbiol. Let.** (1983); [3] Van Kuijk et al., **Anim. Feed Sci. Technol**. (2015); [4] Dorado et al. **App. Microbiol. Biotech**. (2001)

Background

Previous selection for high potential fungi

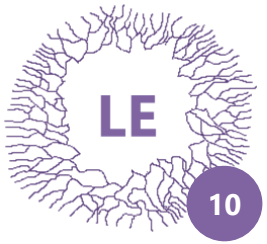
Ceriporiopsis subvermispora

No fruiting body formation



Pleurotus eryngii

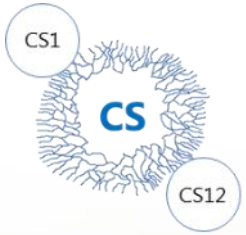
King oyster/trumpet mushroom

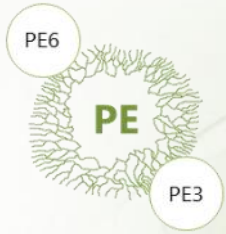


Lentinula edodes

Shiitake mushroom

Selected based on IVGP

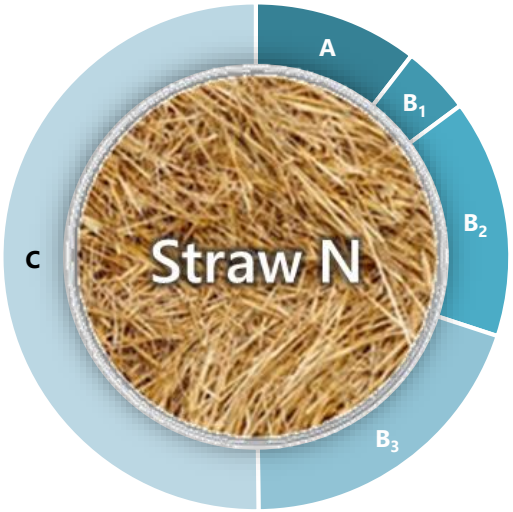




Nayan et al., J. Appl. Microbiol. (2018)

Objective

Fractionation of crude protein [1]



In vitro N availability [2]

[1] Sniffen et al. J. Anim. Sci. (1992); [2] Cone et al., Anim. Feed Sci. Technol. (2009)

Methodology

Fungal preparation

Agar plate

2-3 weeks

Spawn

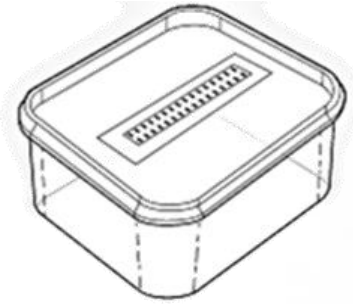
4-5 weeks

Wheat straw preparation

Soaking — 3 days

Autoclave — 1 hour

Treatment



Inoculate wheat straw with spawn (10% dry weight)

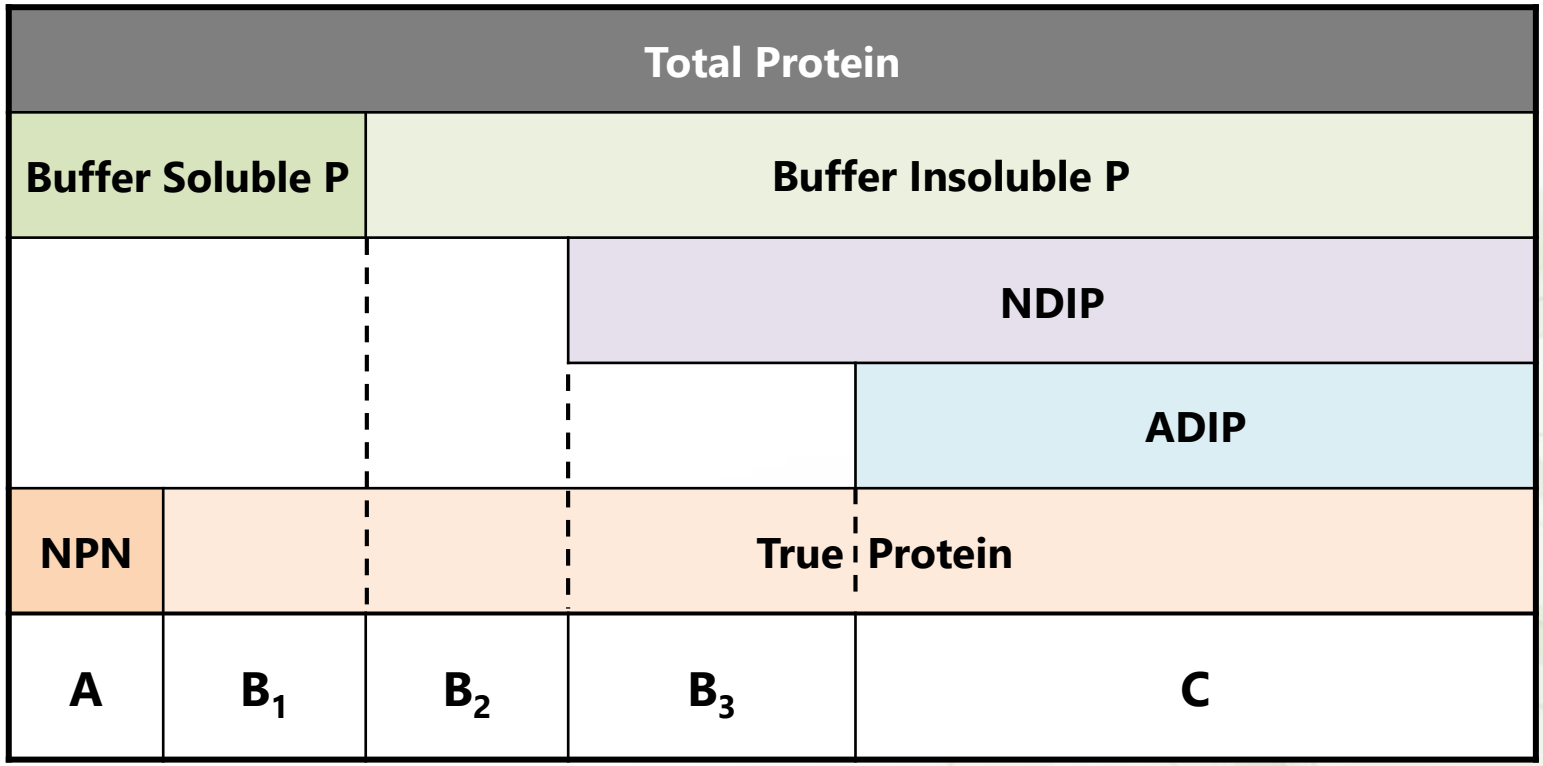
7 weeks

Analyses

- Fractionation of CP
- *In vitro* assessment of N availability
- Amino acids

Methodology

Fractionation of crude protein



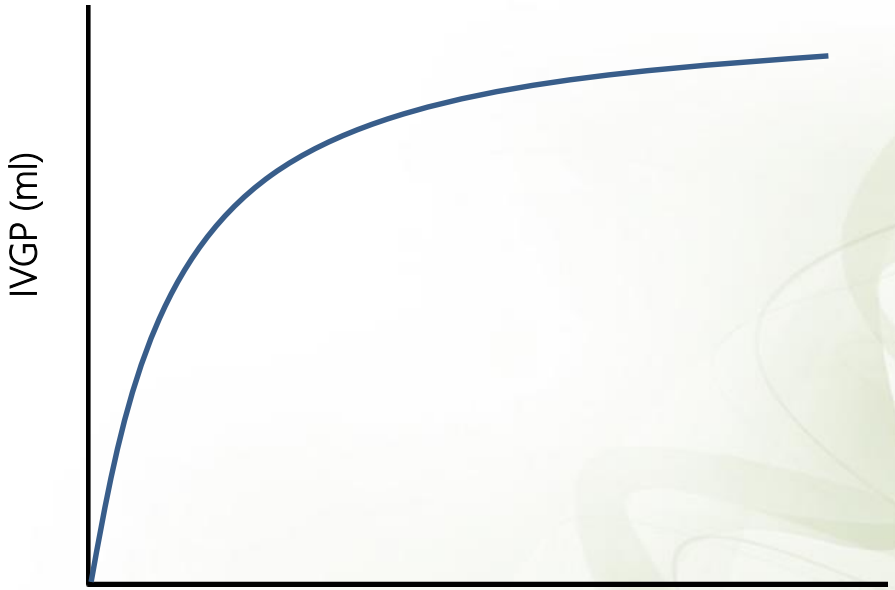
Sniffen et al. J. Anim. Sci. (1992)

Methodology

In vitro assessment of N availability

Rapidly soluble carbohydrates

Pre-incubation (5 h)



Cone et al., **Anim. Feed Sci. Technol.** (2009)

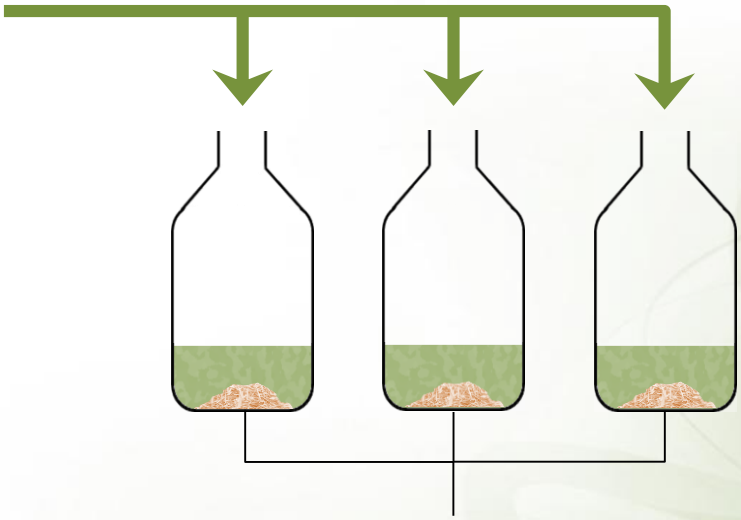
Methodology

In vitro assessment of N availability

Rapidly soluble carbohydrates

Pre-incubation (5 h)

60 ml of pre-incubated rumen fluid + remaining carbohydrates



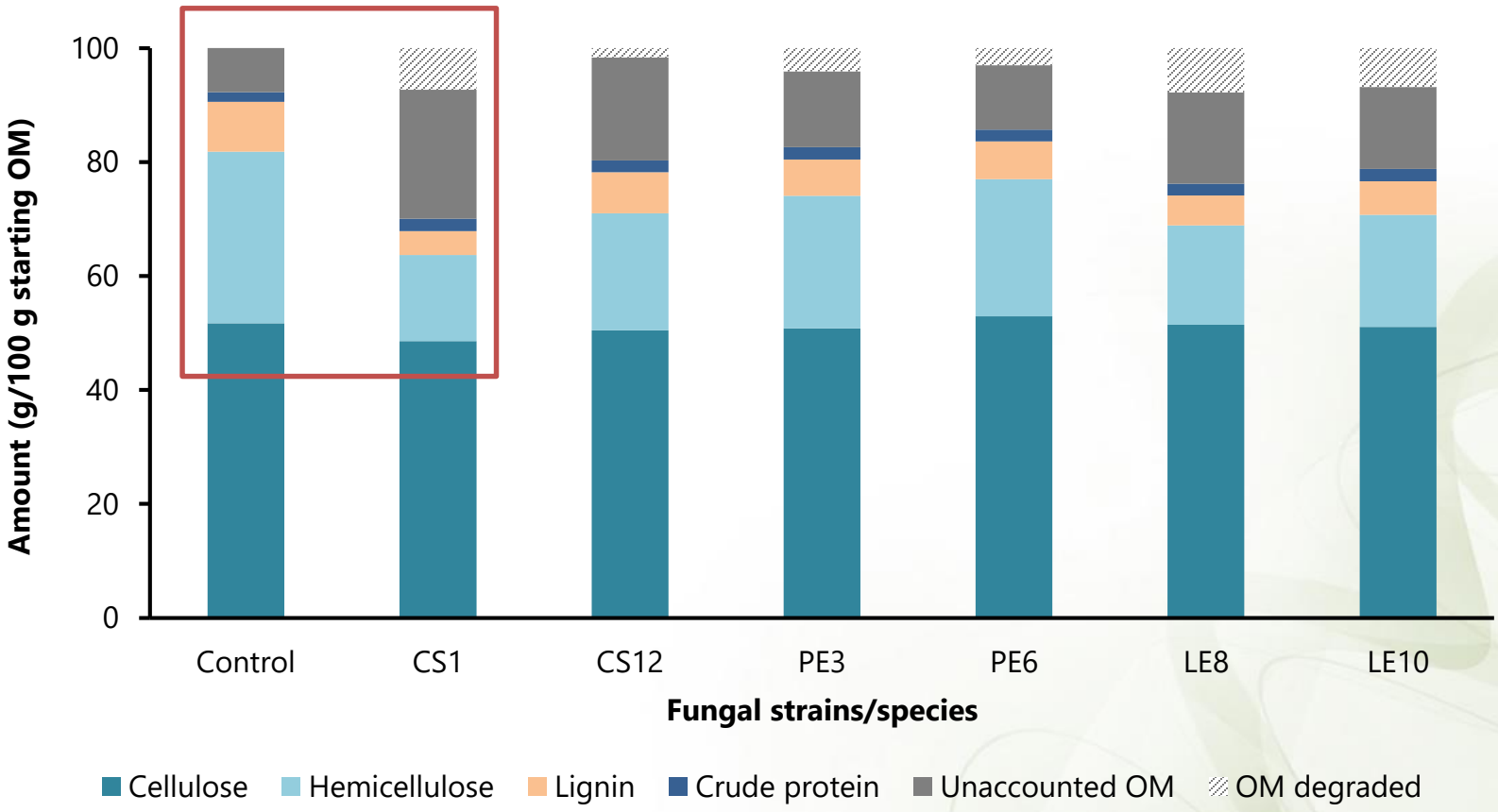
Pre-weighed samples containing 5 mg N

***In vitro* gas production (72 h)**

Cone et al., **Anim. Feed Sci. Technol.** (2009)

Results

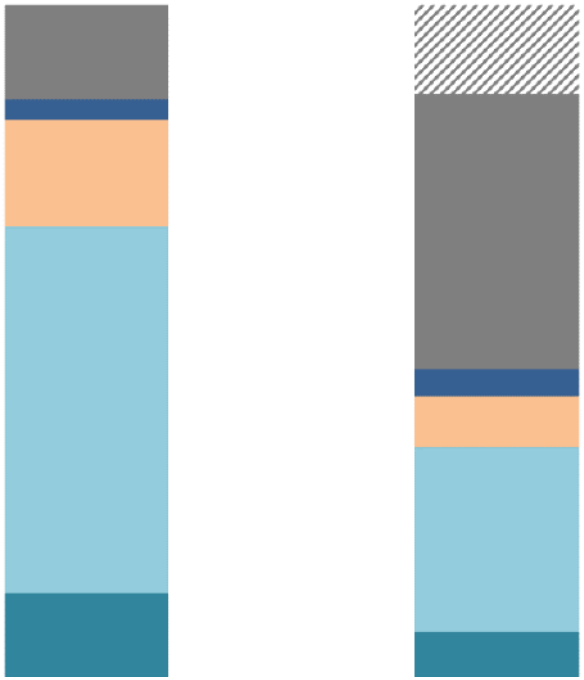
Mass balances



Results

Mass balances

Control vs. CS1-treated wheat straw



Control

CS1

OM degraded ~7.3%

Unaccounted OM 2.9 times higher

CP ↑ 28.2%

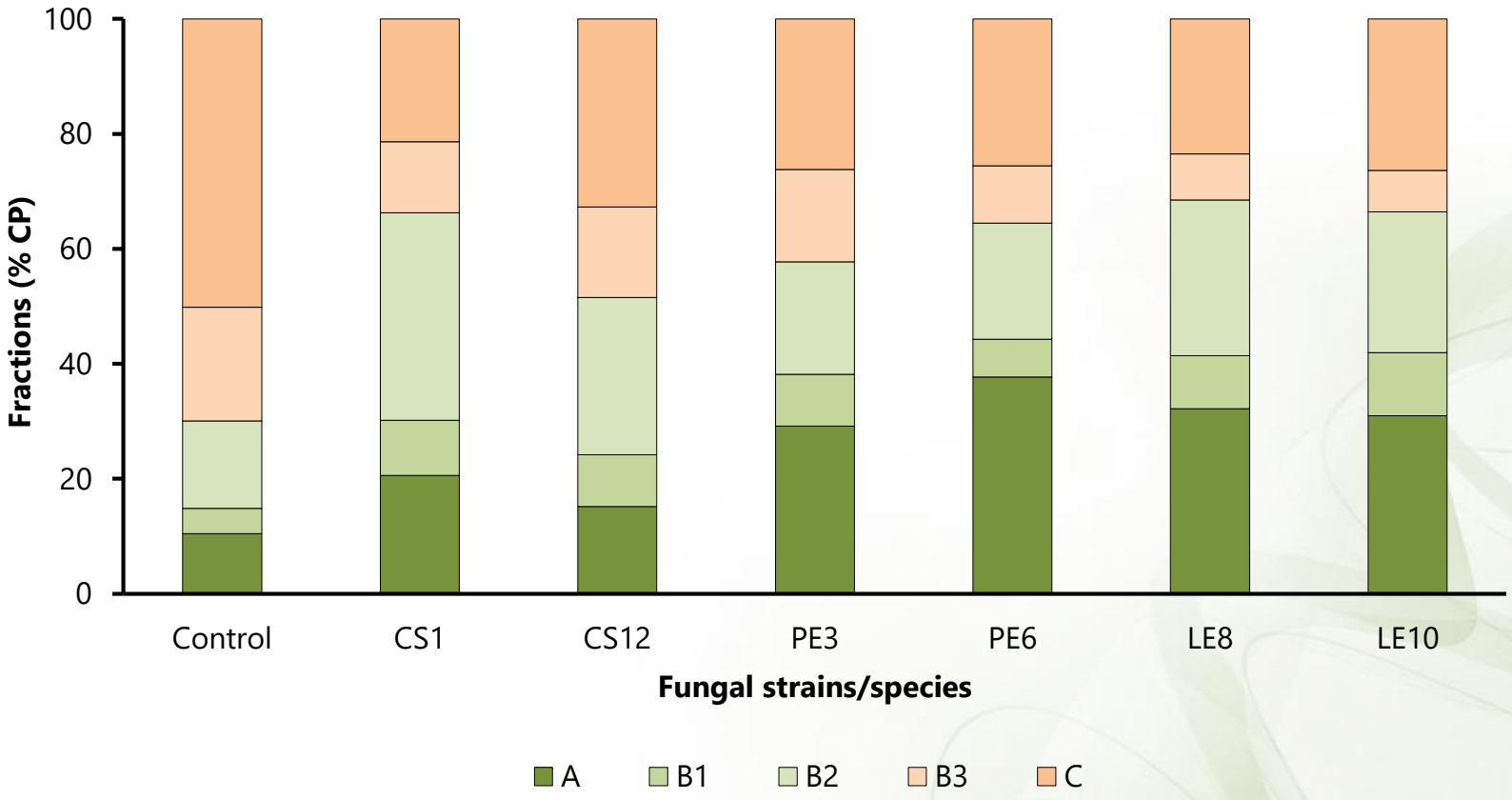
Lignin ↓ 52.2%

Hemicellulose ↓ 49.7%

Cellulose ↓ 6.0%

Results

Fractionation of CP



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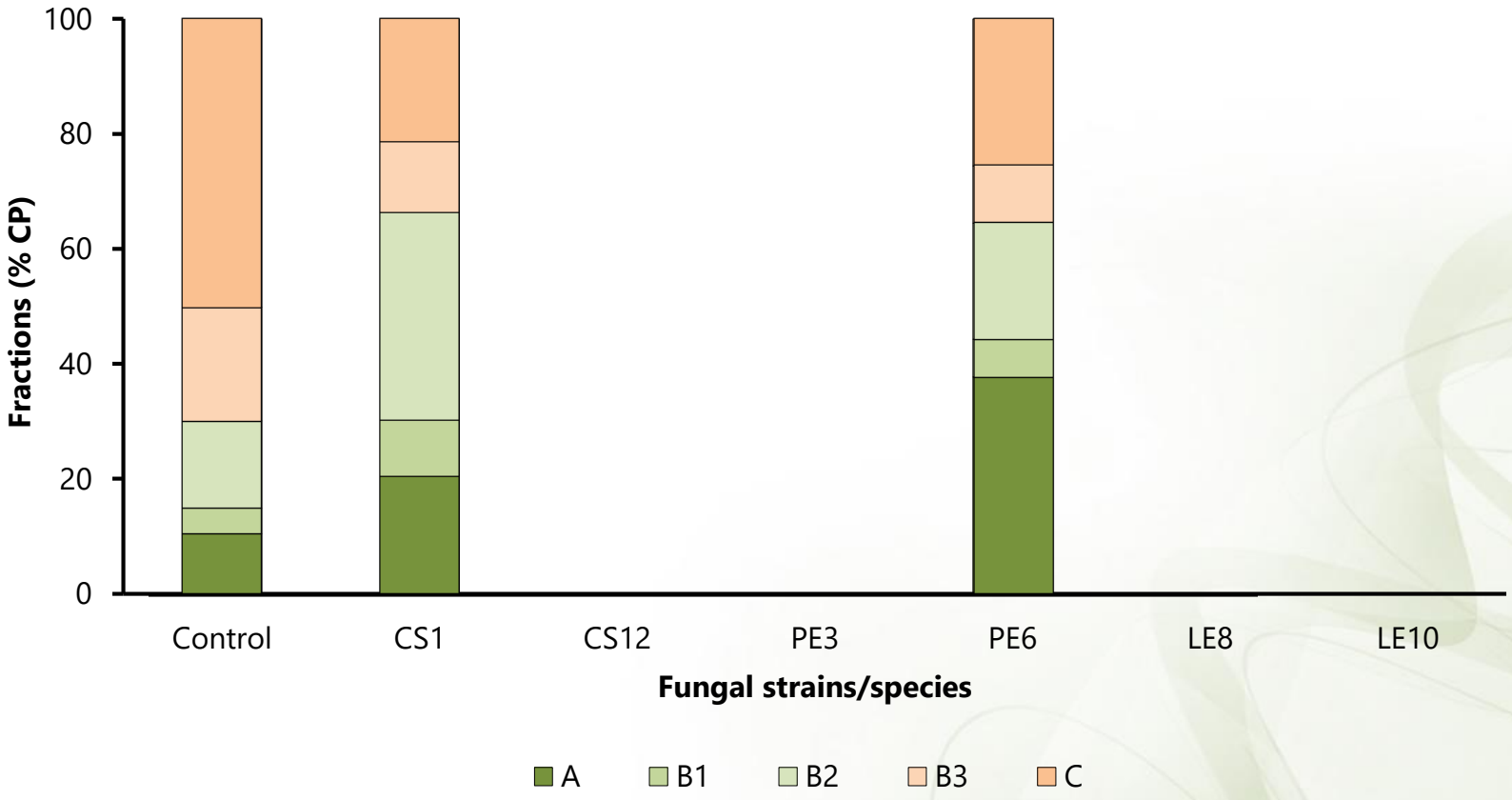

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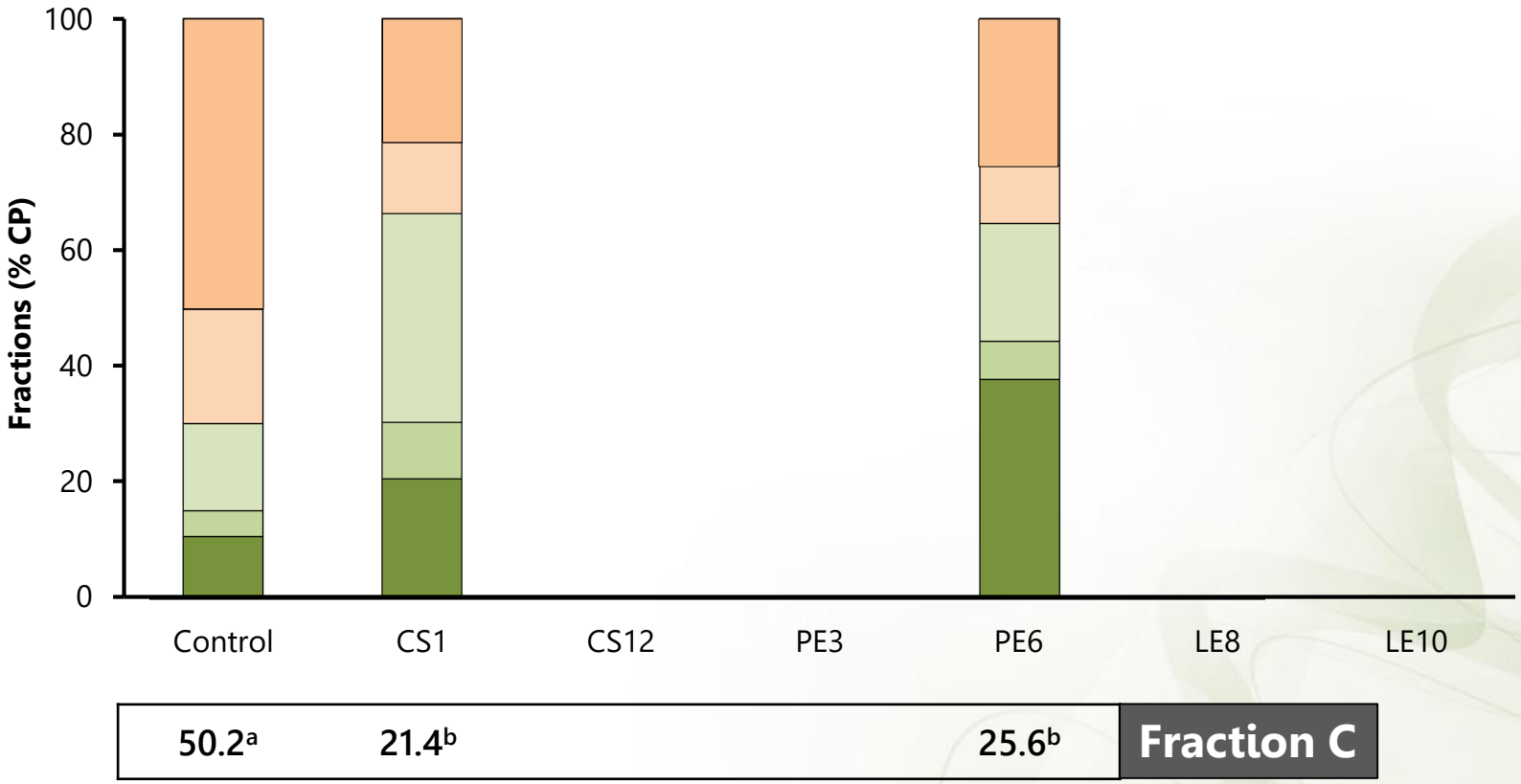
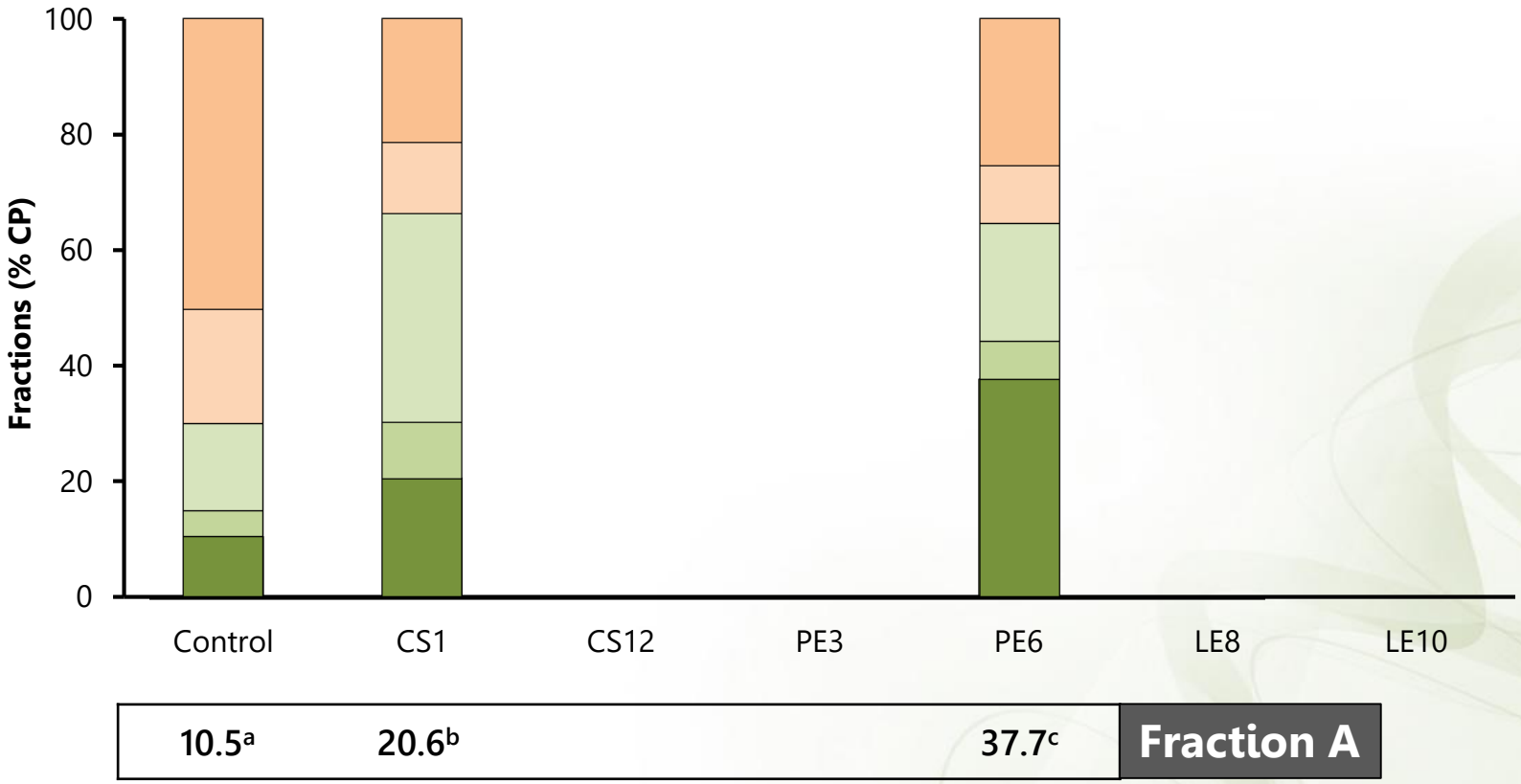
Fractionation of CP



Expressed as %CP. Values with different superscripts are significantly ($P < 0.05$) different

Results

Fractionation of CP



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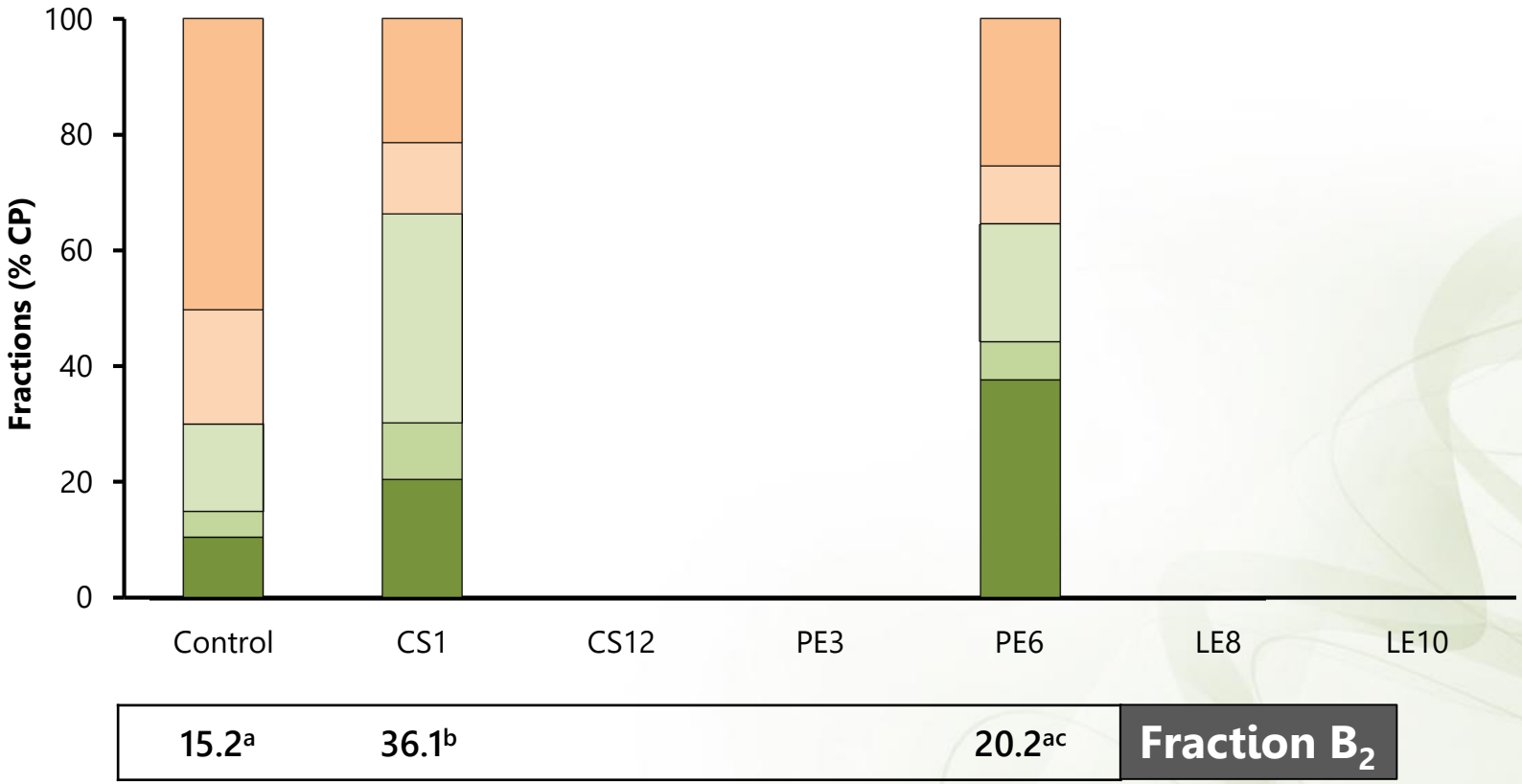
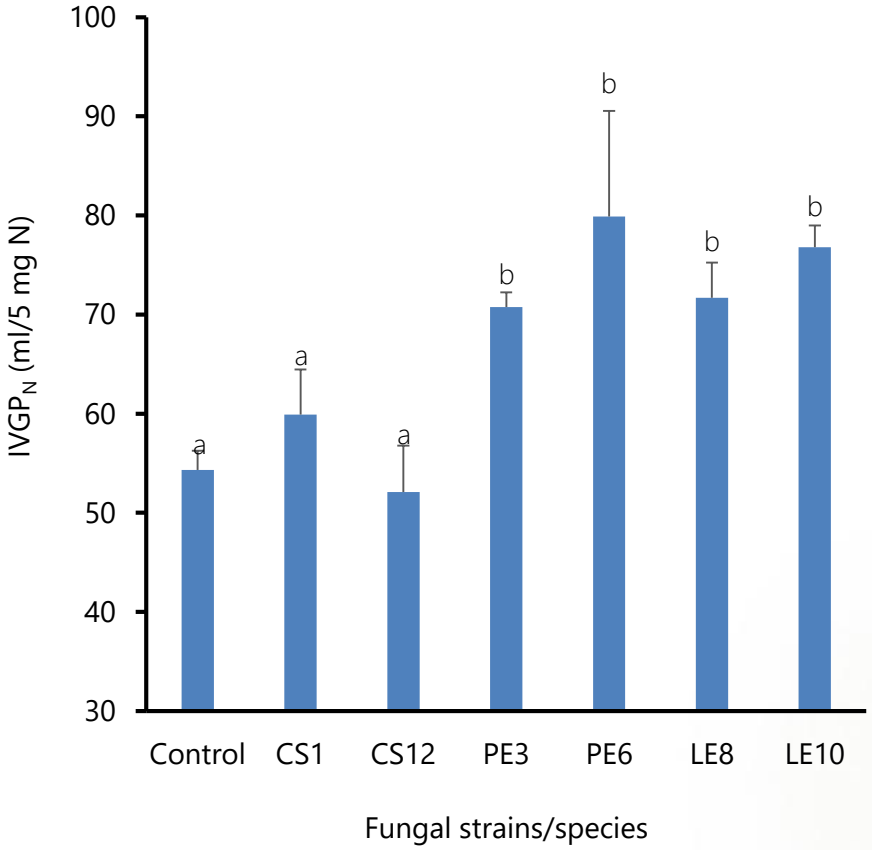


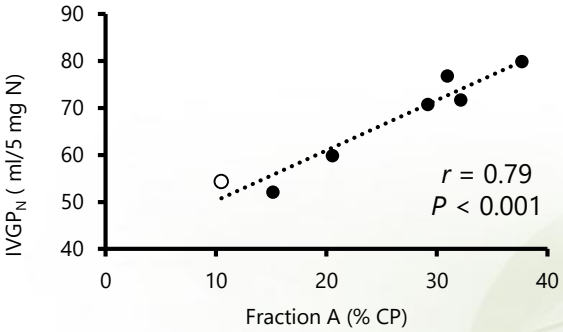
Expressed as %CP. Values with different superscripts are significantly ($P < 0.05$) different

Results

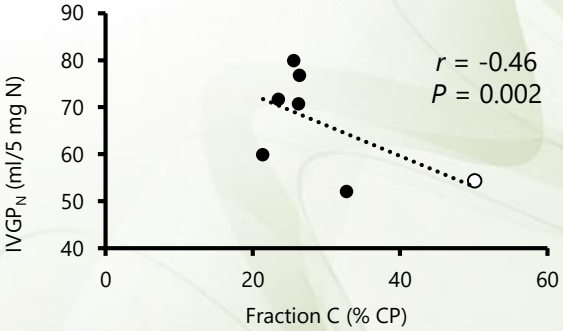
In vitro gas production ($IVGP_N$)



vs. Fraction A



vs. Fraction C



Values with different superscripts are significantly ($P < 0.05$) different

Take-home messages

- Fungi are varied in their capabilities of solubilizing N in the wheat straw
- *P. Eryngii* and *L. edodes* strains increase the N availability in the rumen; but not *C. subvermispora* strains
- Fractionation of protein and *in vitro* evaluation of protein fermentation in rumen fluid are reliable parameters in assessing protein solubilization by different fungal strains.

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Ruminant feed

ABSTRACT

Besides their unique ability to depolymerize cell wall components, white-rot fungi are known to assimilate nitrogenous compounds from substrates. This modification may change protein solubility and fermentation in the rumen. To investigate this, the crude protein (CP) in fungal treated wheat straw (3 fungal species, 2 strains each) was fractioned according to the Cornell Net Carbohydrate and Protein System (CNCPS) and assessed for *in vitro* protein fermentation using a modified gas production technique ($IVGP_N$). Results showed that fungi increased fraction A (instantaneously soluble CP; ~2.6 times) and B_1 (rapidly degradable; ~1.2 times); and decreased the slowly degradable fraction B_3 (~41.6%) and unavailable fraction C (~48.3%). The $IVGP_N$ of straw treated with *Ceriporiopsis subvermispora* strains were not different to the control, but increased by 30.2 to 47.1% in *Pleurotus eryngii* and *Lentinula edodes* strains. The $IVGP_N$ was significantly ($P < 0.01$) correlated to all fractions of CP, except fraction B_1 and B_2 (intermediately degradable). All fungi also increased the arginine (~56%) and lysine (~15%) contents. This study shows the importance of assessing the protein solubilization by different fungal strains, which can uncover unique mechanisms in the cell wall depolymerization.

1. Introduction

In recent years, white-rot fungi such as *Ceriporiopsis subvermispora* and *Lentinula edodes*, have been studied for their ability to improve the degradability of agricultural biomass in ruminants (Tuyen et al., 2013; Van Kuijk et al., 2015a). These reports demonstrate clearly that these fungi are able to modify cell wall properties by selectively degrading lignin and increase the amount of potentially fermentable structural carbohydrates. However, little attention has been paid to the effect of the fungal pretreatment on other nutrients in the biomass, particularly protein. Literature shows an increase of total protein content in fungal-treated biomass (Arora and Sharma, 2011; Sharma and Arora, 2010). Although the protein content in biomass such as wheat straw is low (~16.1 g/kg on dry matter basis) (Nayan et al., 2018), the knowledge on fungal modification of protein availability and solubility is useful and can be applied on various biomasses and even forages. Fungi are known for their capabilities in assimilating nitrogen (N) from the substrate (Davis and Wong, 2010). Although ammonium and glutamine are their preferred nitrogenous compounds, fungi can also use N from various other sources, such as nitrate, urea and amines, to synthesize protein (Tudzynski, 2014). We hypothesize that the modification and assimilation of nitrogenous compounds by fungi may change the solubility and availability of protein in the wheat straw for rumen microbes.

Rumen microbes are able to degrade protein from the feed or directly use ammonia and other non-protein nitrogen (NPN)

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Thank you
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