Nitrogen recycling in relationship with biomass production in a *Miscanthus sinensis* progeny with potential for Ecosystem Services

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Introduction

- Miscanthus, a perennial C4 grass, thrives on marginal lands and offers ecological benefits, with high biomass (Arnoult et al., 2015) making it a promising option for renewable energy (Heaton et al., 2008).
- It also provides ecosystem services (water and soil protection, carbon sequestration, phytostabilisation…) (Nsanganwimana et al., 2014).
- Its efficient nitrogen recycling allows no need for nitrogen fertilization minimizing its environmental impact (Cadoux et al., 2014) and can be beneficial for water and soil protection.
- Most of the cultivated area in Europe is grown with a single clone of *Miscanthus × giganteus.*

  It represents a risk in the slight event of climatic or phytosanitary hazards.
- It has narrow genetic background.

  Hence, we need to diversify the varietal offer for which ***Miscanthus sinensis*** is a very good alternative.
- **Nitrogen recycling** has been studied in *Miscanthus sinensis* (Leroy et al., 2022), but the variability within the species remains largely unexplored.
- Further research is needed to explore the genetic variability in nitrogen recycling and its relationship to biomass yield, to harness this diversity for developing sustainable, high-yield, low environmental impact genotypes.

Miscanthus sinensis

Huge genetic variability (Sun et al., 2010)

Phytostabilisation (Nsanganwimana et al., 2015)

Better abiotic stress tolerance (Lewandowski et al., 2016)

Intraspecific variability (Zub and Brancourt-Hulmel, 2010)

Able to recycle nitrogen as efficiently as *M. x giganteus* (Leroy et al. 2022)

Objective & Hypothesis

Our aim was to explore the variability of nitrogen recycling in *Miscanthus sinensis* progeny in relationship to biomass production.

- Leroy et al. (2022) highlighted nitrogen recycling differences in two genotypes, leading us to anticipate broader variability and a genotype effect among the studied progeny.
- Leroy et al. (2022) also observed that larger plants tend to have higher autumn remobilization. Similarly, we can hypothesize that higher spring remobilization can serve as an endogenous fertilizer, potentially enhancing biomass production like external nitrogen fertilization (Lee et al., 2017).

Material

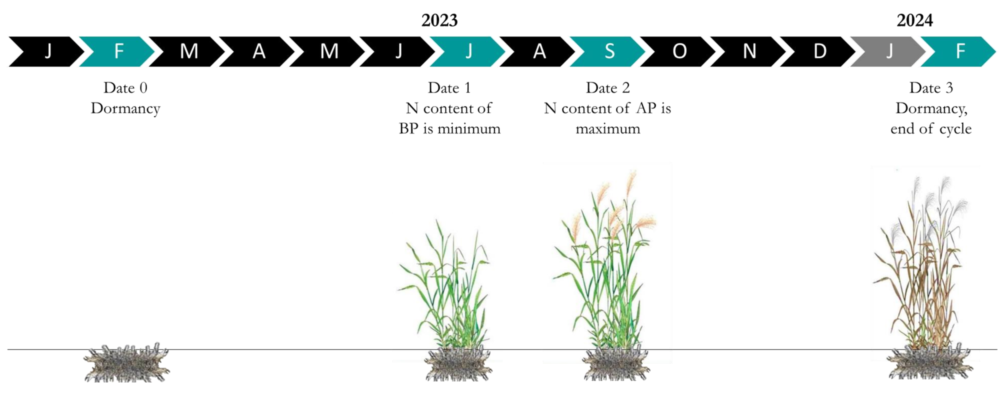
Progeny of a diploid *Miscanthus sinensis* establishedin the field in 2018.

Incomplete block design | 4 blocks | 80 genotypes | 4 or 8 replications, allowing for 1 or 2 plant repetition | 15 repeated genotypes

Methodology

Nitrogen remobilization

- Nitrogen remobilization refers to the **quantity** of nitrogen transferred from belowground parts (BP) to aboveground parts (AP) called spring remobilization (SR), and from AP back to BP called autumn remobilization (AR).
- This involved sampling both AP and BP at four key dates of the growing season.
- Biomass and nitrogen content were characterized to calculate nitrogen fluxes and remobilization efficiencies.



Nitrogen recycling efficiencies

- **Nitrogen remobilization efficiency** is the **proportion** of the total nitrogen in the plant that comes from remobilization (from Dierking et al. 2016).
- It allows to **compare functioning** of different plants.

$$= \frac{\text{Nitrogen remobilized in the period}}{\text{Maximum nitrogen in AP or BP}}$$

Statistical Analysis

- Analyses were conducted using R software V4.3.1.
- ANOVA (car package) was used to assess genotype effect in nitrogen recycling efficiencies.
- Data was log-transformed to achieve homogeneity of variance.
- **LSD (Least Significant Difference)** test was applied (agricolae package) to determine the minimum statistically significant difference between genotypes.

References

Arnoult et al. (2015) https://doi.org/10.1007/s12155-014-9524-7

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Lewandowski (2016) https://doi.org/10.3389/fpls.2016.01620

Sun et al. (2010) https://doi.org/10.1111/j.1095-8339.2010.01082.x

Heaton et al. (2008) https://doi.org/10.1111/j.1365-2486.2008.01662.x

Lee et al. (2017) https://doi.org/10.3389/fpls.2017.00544

Results & Discussion

(1) Large range of nitrogen recycling efficiency is observed across *Miscanthus sinensis* progeny

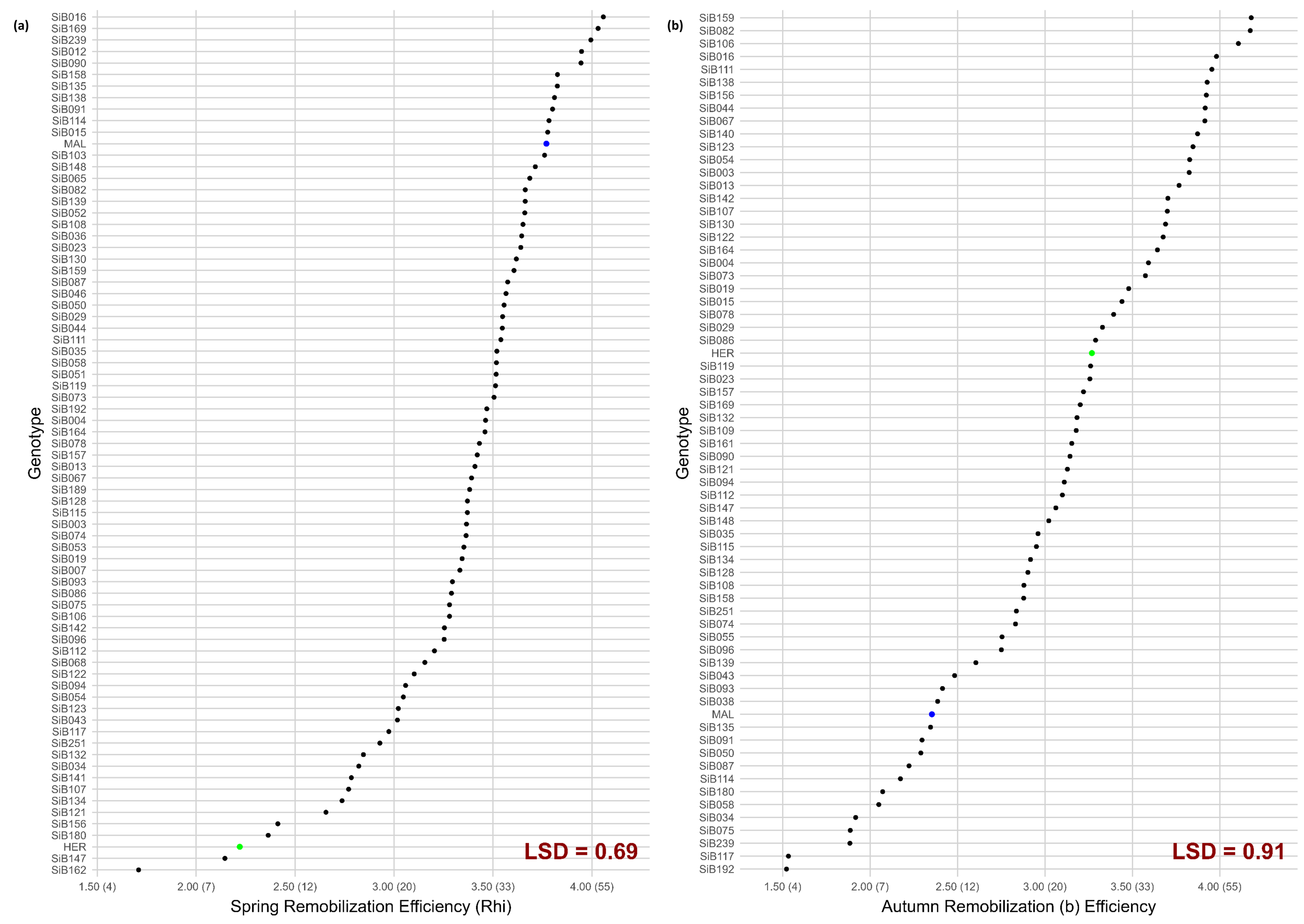

Fig. 1. Spring remobilization efficiency in proportion to maximum nitrogen in BP **(a)** and autumn remobilization efficiency in proportion to maximum nitrogen in AP **(b)** across *Miscanthus sinensis* progeny. The x-axis shows log-transformed values, with corresponding non-transformed values (%) in parentheses. Each point represents the mean value. 'Rhi' and 'b' denote measurements using the belowground method.

- We observed large variability in spring (7% to 58%) and autumn (4% to 65%) remobilization efficiencies, aligning with Leroy et al. (2022), who reported similar ranges (spring: 13% to 56%, autumn: 18% to 37%) in two *M. sinensis* genotypes.
- ANOVA and LSD tests, performed on repeated genotypes, confirmed a significant genotype effect ($p < 0.05$) on nitrogen recycling efficiency in spring and autumn. LSD value indicates the smallest difference needed to distinguish genotypes statistically (Fig 1).

(2) Nitrogen recycling efficiency does not guarantee higher biomass yield and vice versa at inter-genotypic level

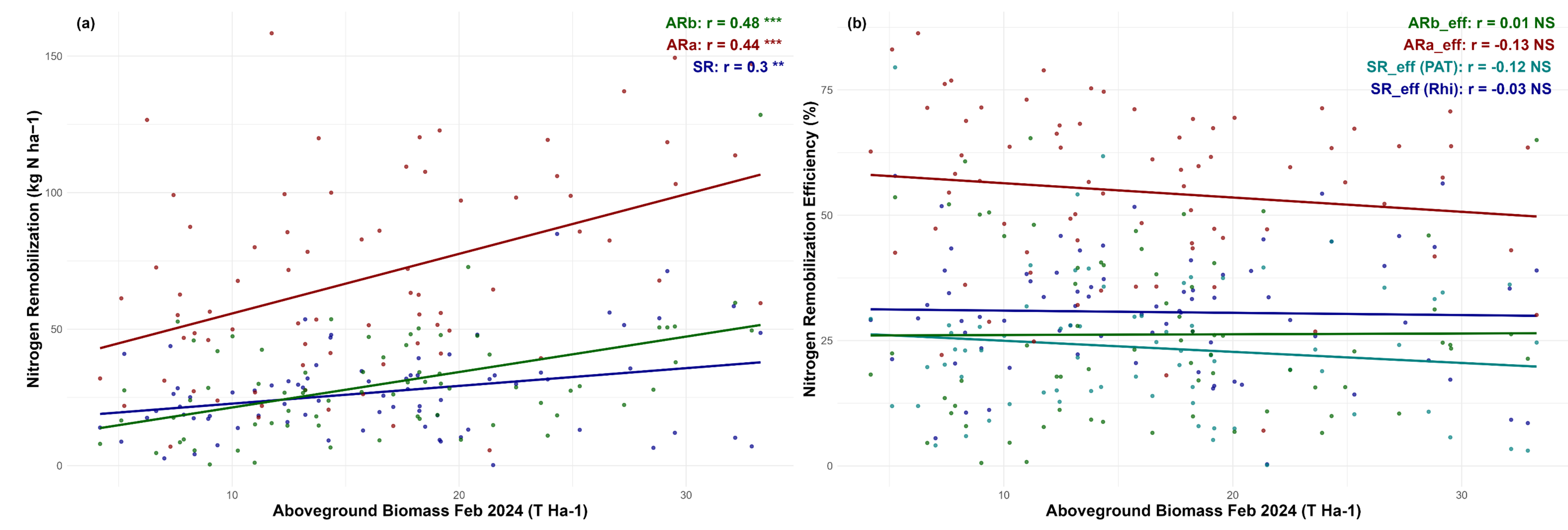

Fig 2. Relationship between nitrogen remobilization quantities **(a)** and nitrogen remobilization efficiencies **(b)** with biomass yield. The remobilization quantities are SR (Spring Remobilization), ARa (Autumn Remobilization measured using the aboveground method), and ARb (Autumn Remobilization measured using the belowground method), along with their respective efficiencies. Pearson correlation coefficients (r) are shown, with *** for $p < 0.001$ and NS for non-significant correlations ($p \geq 0.1$).

- Despite the significant relationship between remobilizations and biomass (Fig. 2a), the dispersion among genotypes weakens these correlations. A more consistent relationship could be expected at the intra-genotypic level.
- Lack of correlation between remobilization efficiencies and biomass (Fig. 2b) suggests that plant functioning, in terms of nitrogen recycling, operates independently of biomass size.

Conclusion & Perspectives

- The observed variability in nitrogen recycling among *M. sinensis* progeny encourages future genetic studies (heritability, QTL detection). Further research is in progress at intragenotype level with more replications with few genotypes.
- Our study will continue to explore the relationship between nitrogen recycling, nitrogen use efficiency (NUE) and external nitrogen uptake to better characterize plant nitrogen functioning.
- As the plant nitrogen functioning operates independently of biomass production,

  It allows us in a next step to integrate nitrogen recycling and biomass production to define different functional groups which can be interesting for different ecosystem services. For example;

  Low nitrogen recycling, high biomass genotypes can be interesting in water catchment areas polluted by nitrates.

  High nitrogen recycling, high biomass can be interesting in lands polluted by heavy metals or low input areas.

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Biomass and Energy Crops VI Conference
1st - 3rd October 2024
Aberystwyth University - WALES