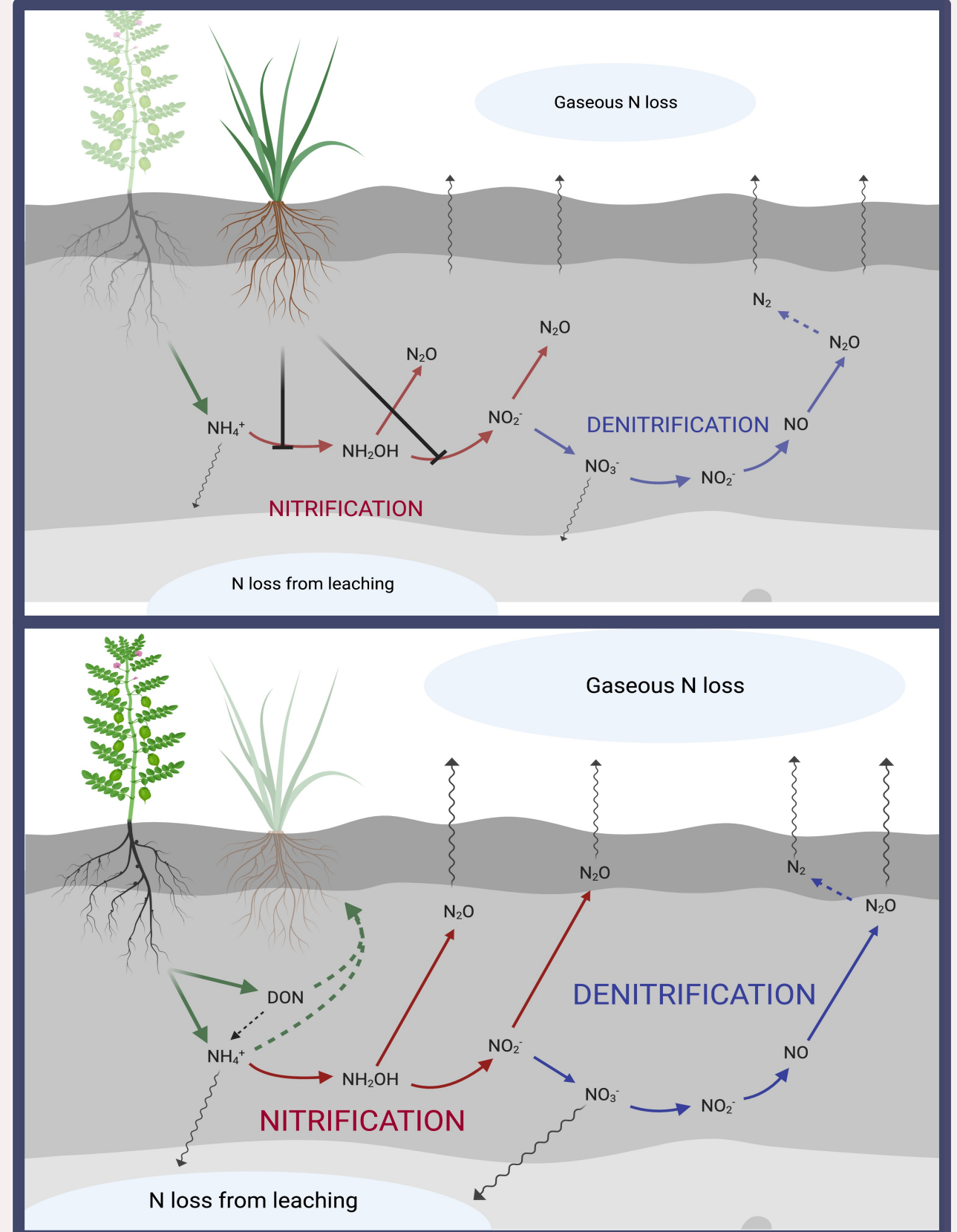


## Introduction



*Brachiaria* prevents potential N loss through biological nitrification inhibition (BNI). Intensive harvesting can result in yield declines in unfertilized pastures.



### Expected Results

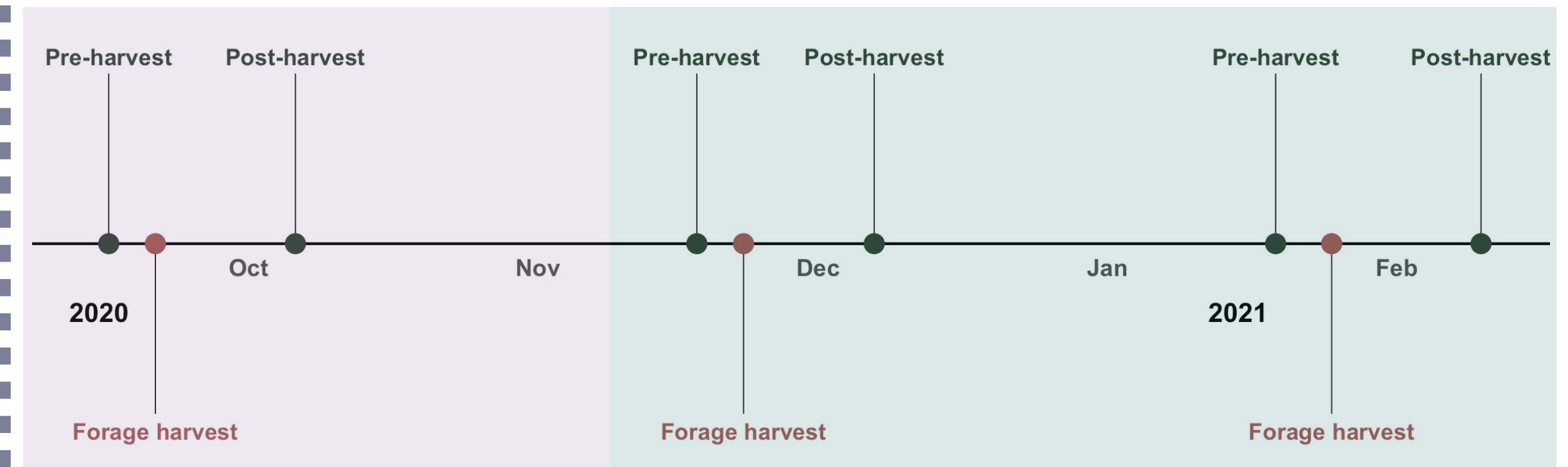
- Brachiaria* will limit potential sources of N-loss by lowering nitrifier and denitrifier enzyme activity relative to the baseline (*C. purpureus*). Perennial crops will also exhibit lower NP and DEA relative to farmer-preferred annual maize.
- A legume intercrop will increase NP and DEA compared to monocrop treatments. DEA will increase in the rainy season and NP activity will be greatest in the dry season.

## Methods

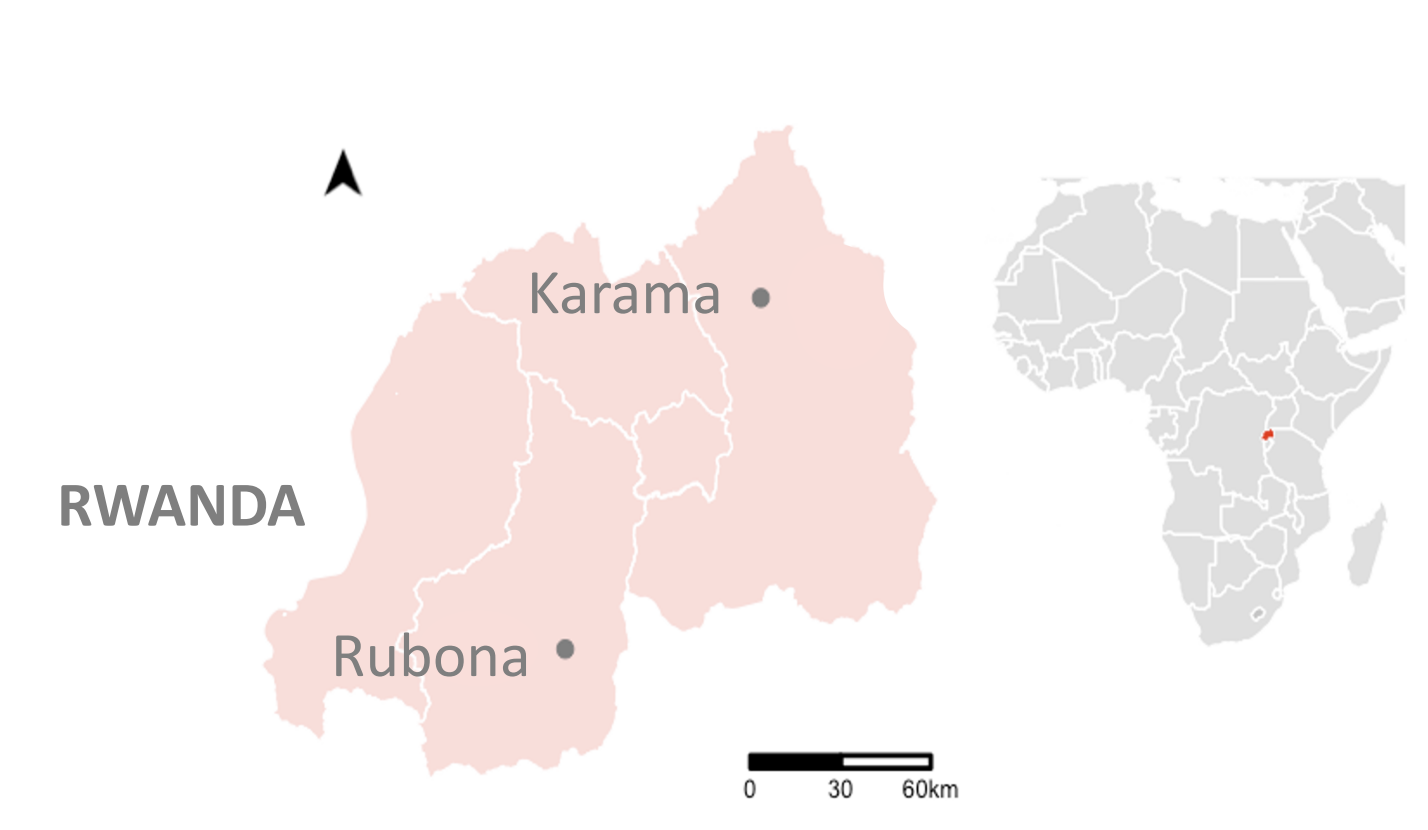
### Forage Treatments



### Soil Sampling



### Study Locations



Forage trials were planted in October 2019 and harvested every ~8 weeks since establishment. Maize was not harvested during the course of data collection.

Bulk soil was collected to a depth of 12cm after removing the top layer of leaf litter. Soil was collected six times at each location between September 2020 and March 2021. These sampling events spanned both dry and rainy seasons, occurring immediately before and two weeks following forage harvest to account for contrasting periods of crop N demand.

### Soil Analyses

**NP**  
 1. 2.5 g soil, 25 ml NP solution (K<sub>2</sub>HPO<sub>4</sub>, (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>)  
 2. 36 hr incubation on orbital shaker (180 rpm)  
 3. Collect 1.25 ml samples  
 4. Centrifuge  
 5. Quantify NO<sub>3</sub><sup>-</sup>  
 Timers: 2 hrs, 12 hrs, 24 hrs, 36 hrs

**DEA**  
 1. 2.5 g soil, 5 ml DEA solution (KNO<sub>3</sub>, D-glucose)  
 2. Evacuate vials with gas manifold, Add C<sub>2</sub>H<sub>2</sub>  
 3. 90 min incubation on orbital shaker (180 rpm)  
 4. Sample headspace  
 5. Analyze gas samples on GC-MS  
 Timers: 0 min, 45 min, 90 min

## Research Questions

- Does the climate-smart forage *Brachiaria* reduce potential N loss from microbial pathways compared to preferentially grown non-BNI forage crops such as Napier grass (*Cenchrus purpureus*) or annual maize (*Zea mays*)? To what extent is this effect mediated by season?
- Are nitrification and denitrification stimulated by the presence of a perennial forage legume (*Desmodium*) intercrop?

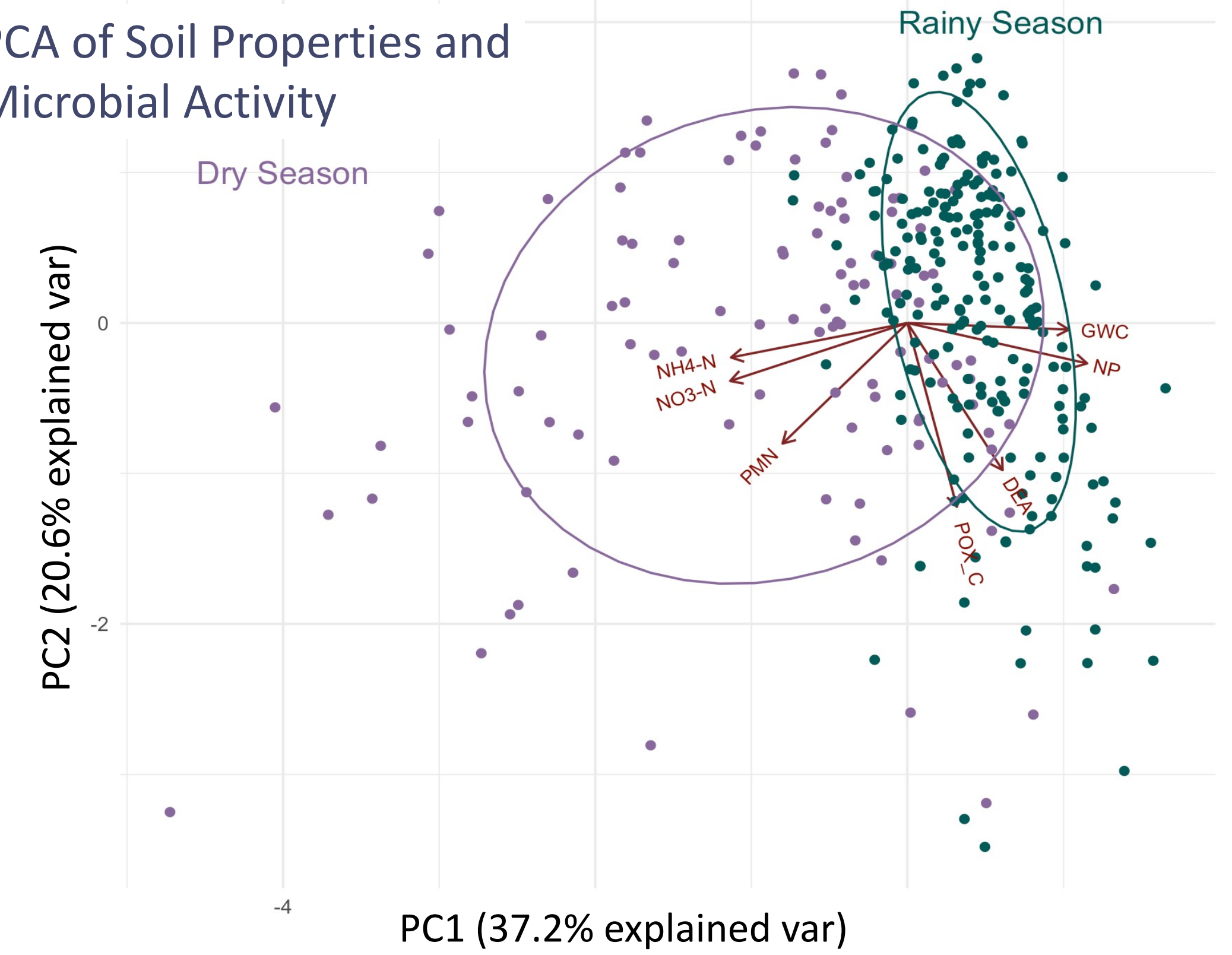
## Results

Karama: Nitrification Potential		Rubona: Nitrification Potential	
Treatment	Predicted	Treatment	Predicted
Napier Grass	~1.5	Napier Grass	~1.5
Brachiaria cv. Mulato II	~1.5	Brachiaria cv. Mulato II	~1.5
Annual Maize	~1.5	Annual Maize	~1.5
Napier + Desmodium	~1.5	Napier + Desmodium	~1.5
Brachiaria + Desmodium	~1.5	Brachiaria + Desmodium	~1.5
Maize + Desmodium	~1.5	Maize + Desmodium	~1.5
Desmodium distortum	~1.5	Desmodium distortum	~1.5

Predicted value is defined as the estimated marginal means of treatment effects from a mixed effects linear model. Treatment and collection timepoint were both treated as fixed effects, with block and timepoint as random effects. Dashed vertical line: marginal mean effect of the Napier grass (*C. purpureus*) monocrop treatment, which as treated as the control group.

- Brachiaria* did not significantly decrease NP in either location compared to Napier (*C. purpureus*).
- Maize tended to have lower nitrification (NP) (Rubona: p<.05; Karama: ns) and denitrification (DEA) potential activity compared to Napier (Rubona: p<.05; Karama: ns).
- Legume intercropping did not increase NP or DEA relative to monocrop treatments.
- Forage growth stage had a significant effect on NP in the rainy season (Rubona & Karama: p<.001) but not the dry season (Rubona & Karama: ns).

### PCA of Soil Properties and Microbial Activity



- Soil moisture was a strong driver of N dynamics and microbial activity, regardless of location or planting treatment
- NP and DEA increased with soil gravimetric water content (r=0.32, p<.001; r=0.2, p<.001), even as putative leaching in the rainy season decreased min-N availability

## Conclusion

Maize plots had low nitrification and denitrification activity compared to *Brachiaria* and Napier plots; this may be due to root-mediated effects on microbial activity from intensive forage harvesting<sup>2</sup>. Legume intercrops did not stimulate NP or DEA compared to monocrops, suggesting that they can provide N inputs without contributing to N loss. Intercropping BNI forage grasses with perennial legumes is a novel strategy to provide both N-retention and N-provision services in tropical agriculture. However, additional strategies beyond BNI crops may be necessary to conserve N during the rainy season in highly weathered soils.

## Acknowledgements

This research was conducted as part of the CGIAR Research Program on Livestock and is supported by CGIAR Fund Donors. Additional support was provided by the National Science Foundation Graduate Research Fellowship Program (no. 00074041). We are grateful for support from our colleagues in the Grossman Lab: Dr. Vivian Wauters, Dr. Adria Fernandez, Sarah Duber, Tanner Beckstrom, and Bonsa Mohammed. This work was made possible by a dedicated field team in Rwanda led by Paulin Mutanguha and Jean-Claude Majuga. Finally, we thank CIAT office staff in Kenya and Rwanda for managing research-associated logistics.

## References

- Zhao, M., Jones, C.M., Meijer, J., Lundquist, P.O., Fransson, P., Carlsson, G., and Hallin, S. (2017). Intercropping affects genetic potential for inorganic nitrogen cycling by root-associated microorganisms in *Medicago sativa* and *Dactylis glomerata*. *Appl. Soil Ecol.* 119, 260–266.
- Hamilton, E.W., Frank, D.A., Hinchey, P.M., and Murray, T.R. (2008). Defoliation induces root exudation and triggers positive rhizospheric feedbacks in a temperate grassland. *Soil Biol. Biochem.* 40, 2865–2873.