

Elisabeth Diatta-Holgate^{1*}, Cyril Diatta², Clifford Weil³, Ndiaga Cisse¹, and Mitch Tuinstra³

¹ Senegalese Agricultural Research Institute – Regional Center of Research on the Improvement of Plant Adaptation to Drought (CERAAS), Senegal

² *Senegalese Agricultural Research Institute – National Center of Agronomic Research (CNRA), Bambey, Senegal*

³ Agronomy Department, Purdue University, West Lafayette, IN 47907, USA

*Contact Information: ediatta@purdue.edu / lisbethdiatta@yahoo.fr



Introduction

Sorghum is a staple crop and a major source of food and energy for developing countries in Africa and Asia. However, its use as human food is constrained by the low digestibility of its proteins after wet cooking¹, which could contribute to malnutrition. Limited natural variability exists for protein digestibility hence the need to explore allelic variants in induced mutants to improve elite sorghum lines consumed as food. In Senegal, breeding efforts have been made to improve the digestibility of elite white grain, tannin-free sorghum varieties with up to 13.62% proteins that were crossed to P721Q, a high lysine-highly digestible mutant. This resulted to 12 BC₃F₆ having yield comparable to the recurrent parent, Faourou, tolerant to grain mold and with up to 31% increase in protein digestibility. This study also revealed that mutations in a disulfide isomerase protein can lead to an increase in the amount of readily digested proteins in sorghum grains after wet cooking.

Objectives

- Identify allelic variants controlling the increase in protein digestibility in sorghum grains and develop genetic markers that can be used for marker assisted selection.
- Improve the post-cooking protein digestibility of sorghum varieties in Senegal and evaluate their yield performance across the country.

Materials and Methods

- F2:3 mapping population, made from a poorly digestible mutant SbEMS932 (P2) crossed to a highly digestible mutant SbEMS1613 (P1), was assayed for digestibility.
- 2 Pairs of tetra primers developed and tested followed by Sanger sequencing.
- 12 BC3F6 (Faourou x P721Q) tested in 5 locations in Senegal.

High Throughput Digestibility Assay: 60 ± 2 mg of seeds were wet ground, cooked at 95°C for 20 min, and then digested with pepsin for 2h at 37°C . The remaining proteins were extracted, precipitated using trichloroacetic acid, and the A_{560} of the sample compared to an undigested control sample of the same, ground material ².

Bulked Segregant Analysis: Candidate genes were identified through the Simple pipeline³ where SNP frequencies of the low and high digestible bulks were compared.

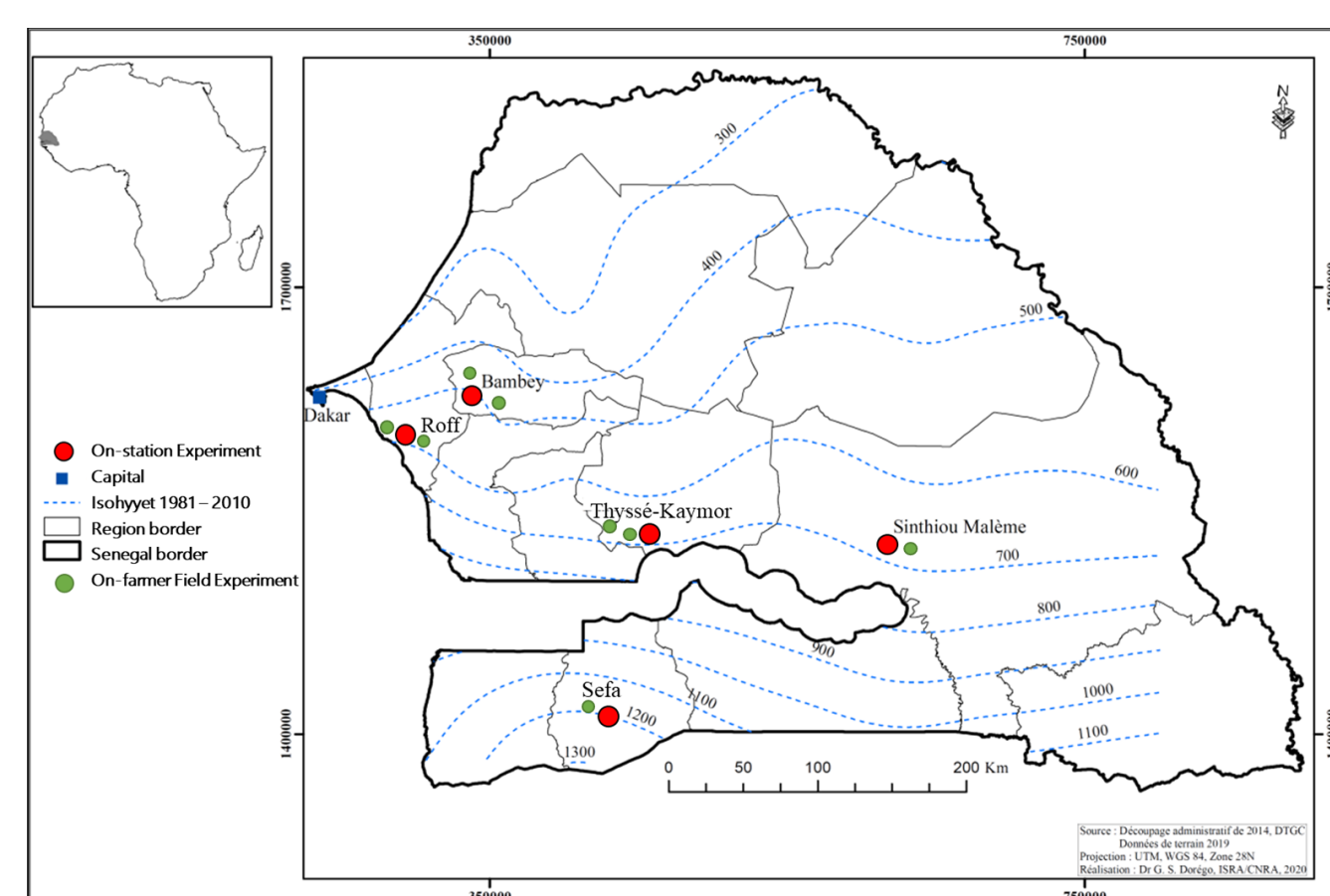


Figure 1: Map of Senegal showing the 5 experimental locations (red dots)



Figure 2: Participatory varietal selection at 2 experimental locations

Results

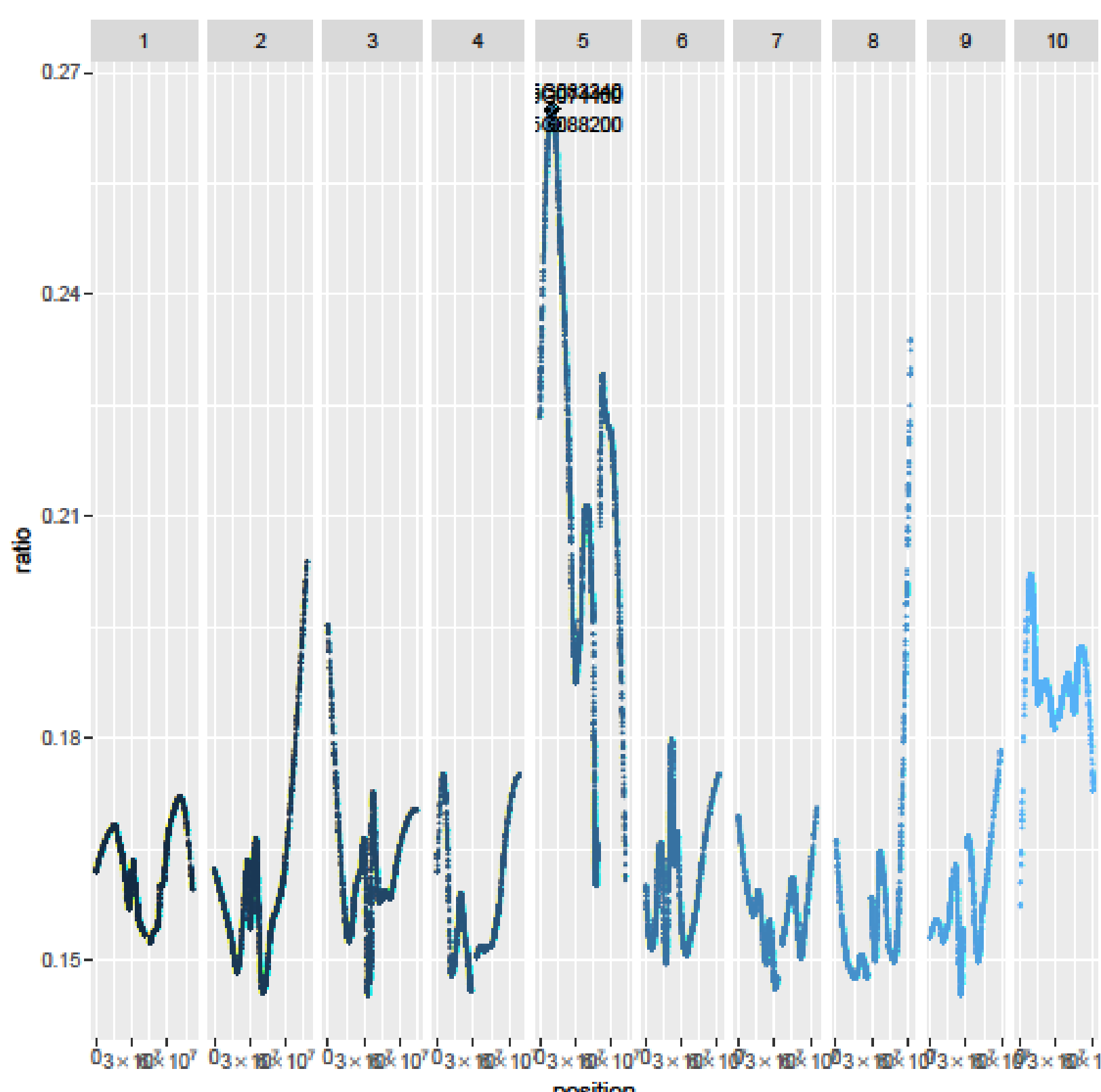


Figure 3: Bulk segregant analysis showing regions on chromosome 5 significantly associated with the increase in digestibility. Three Candidate genes showed on the table.

- 3 candidate genes identified

Chr.*	Candidate Genes
5	SORBI_3005G074400
5	SORBI_3005G083340
5	SORBI_3005G088200
5	SORBI_3005G189000

* Chr: chromosome number

Results

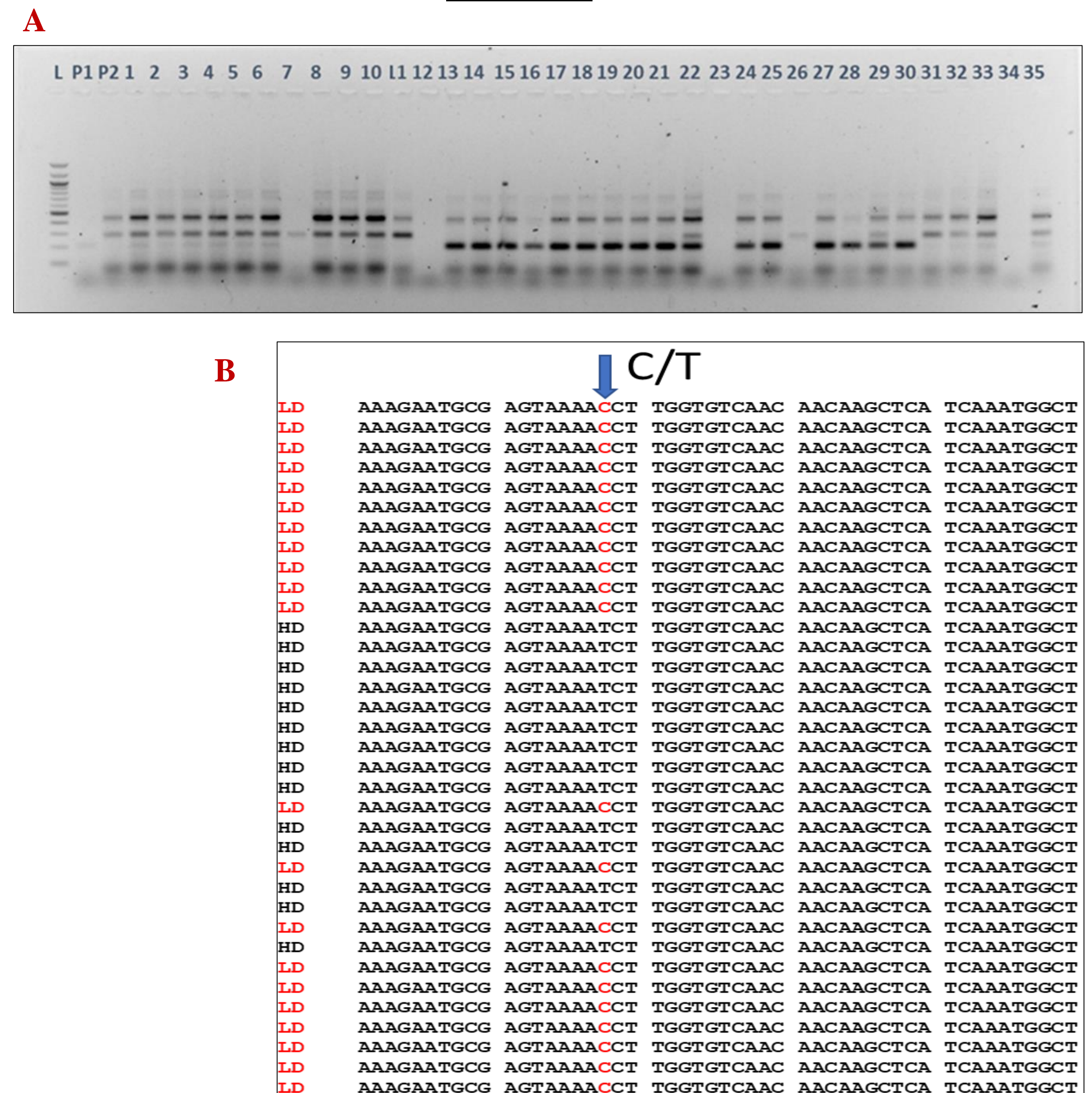


Figure 4: Genotyping results for the Disulfide Isomerase. **A:** Amplification of the isomerase alleles in the high (P1), low (P2) parents and F3 progenies (1-35). L, 100 bp DNA ladder. **B:** Sequence alignment showing that the C/T mutation segregates perfectly with the low (LD) and high (HD) digestible phenotype.

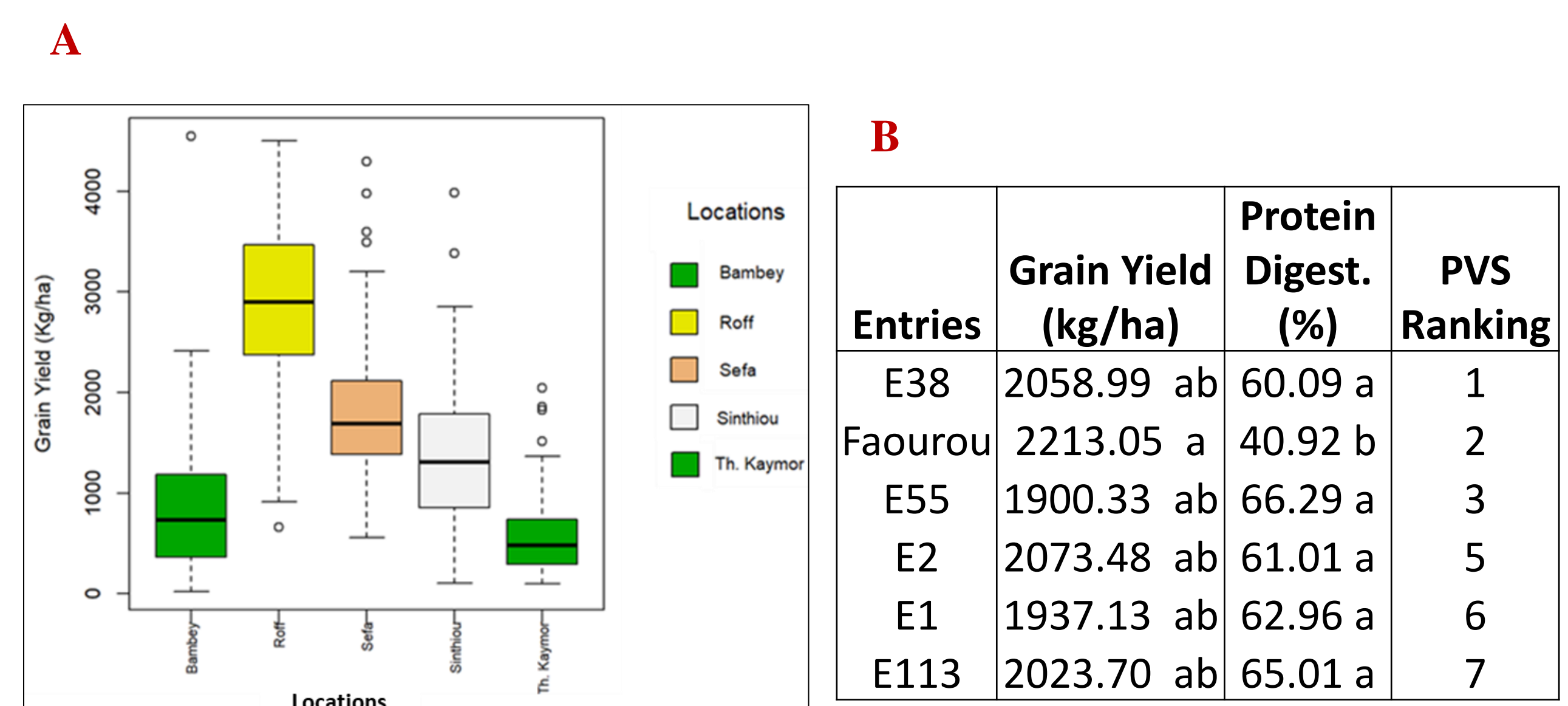


Figure 5: **A:** Yield variation in the 5 experimental locations. **B:** 5 selected best performing lines across locations in terms of yield (comparable to Faourou), protein digestibility (+26%) and farmers ranking. PVS: participatory varietal selection.

Conclusions

- Mutations on the disulfide isomerase protein leads to improved protein digestibility.
- Polymorphic markers will help in marker-assisted selection for protein digestibility.
- Five improved sorghum lines have up to 26% increase in digestibility compared to Faourou.

References

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Acknowledgments

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