Genetic Improvement of Post-cooking Protein Digestibility of Sorghum Grain for Human Consumption

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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 cooking[1], 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 BC_3F_6 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 pipeline[3] where SNP frequencies of the low and high digestible bulks were compared.

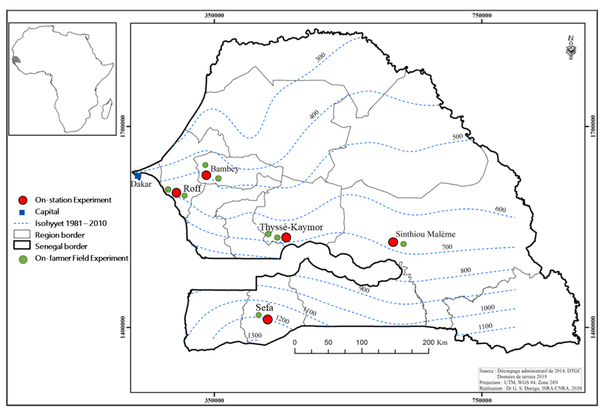
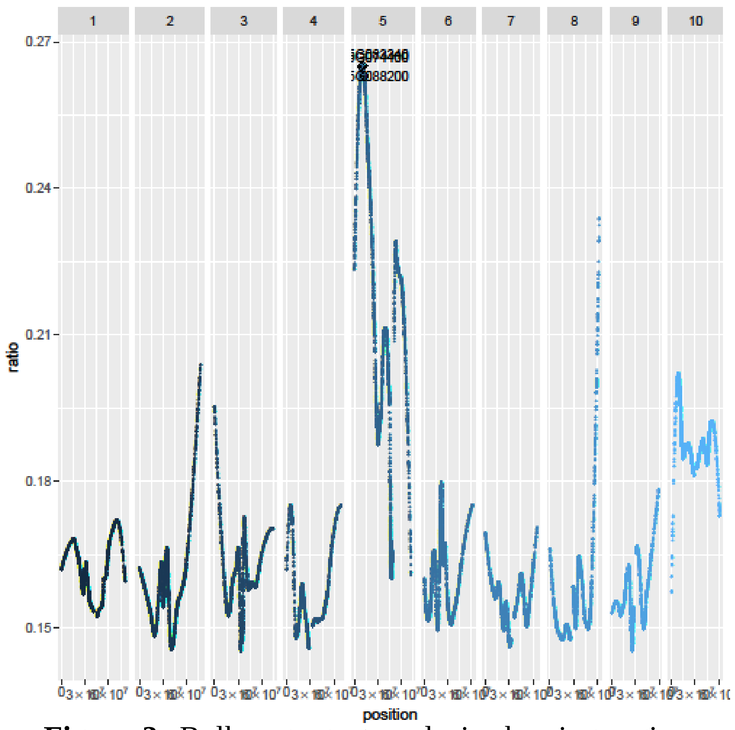


Figure 1: Map of Senegal showing the 5 experimental locations (red dots)

Figure 2: Participatory varietal selection at 2 experimental locations

Results



- 3 candidate genes identified

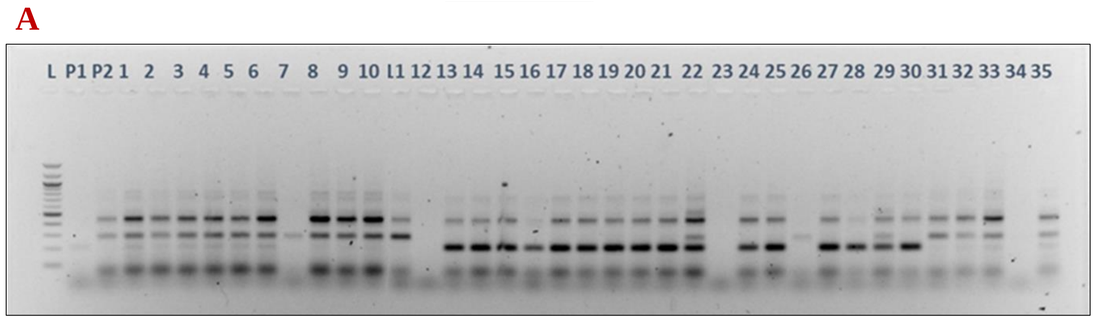
Chr.*	Candidate Genes
5	SORBI_3005G074400
5	SORBI_3005G083340
5	SORBI_3005G088200
5	SORBI_3005G189000

* Chr: chromosome number

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

Results

A



B

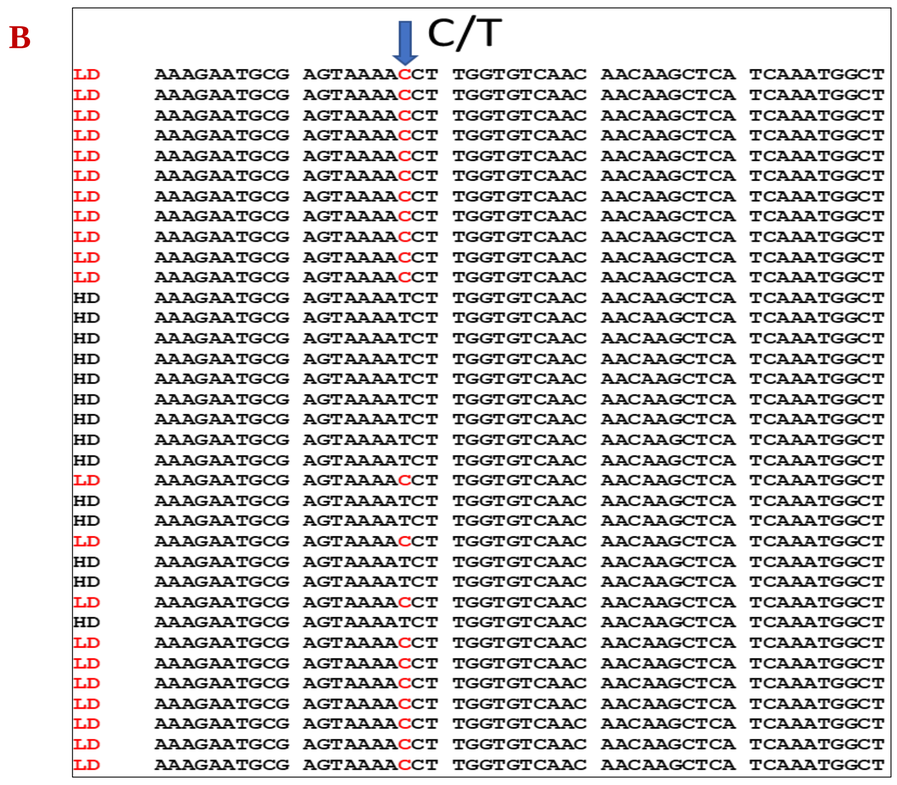
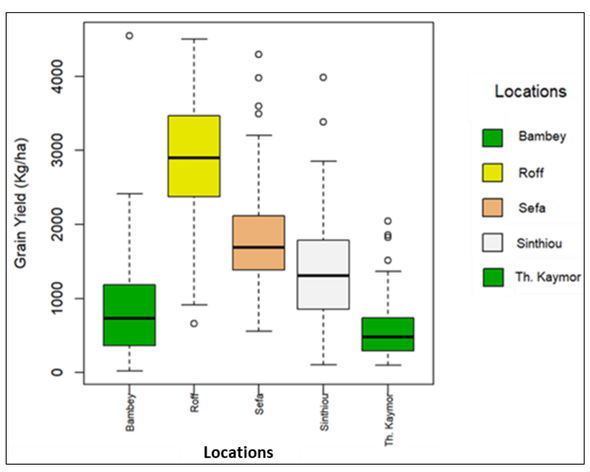


Figure 4: Genotyping results for the Disulfide Isomerase. **A:** Amplification of the isomerase alleles in the high (P1), low (P2) parents and F3 progenies (1-35). L, 100 bp DNA ladder. **B:** Sequence alignment showing that the C/T mutation segregates perfectly with the low (LD) and high (HD) digestible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

A



B

Entries	Grain Yield (kg/ha)	Protein Digest. (%)	PVS Ranking
E38	2058.99 ab	60.09 a	1
Faourou	2213.05 a	40.92 b	2
E55	1900.33 ab	66.29 a	3
E2	2073.48 ab	61.01 a	5
E1	1937.13 ab	62.96 a	6
E113	2023.70 ab	65.01 a	7

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-assister selection for protein digestibility.
- Five improved sorghum lines have up to 26% increase in digestibility compared to Faourou.

References

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Acknowledgments

- This study is made possible through funding by the Feed the Future Innovation Lab for Collaborative Research on Sorghum and Millet through grants from American People provided to the United States Agency for International Development (USAID) under cooperative agreement number AID-OAA-A-13-00047. The contents are the sole responsibility of the authors and do not necessarily reflect the views of USAID or the US Government..
- A special thank you to Claire King, Arushi Arora, Eugene Glover and Andy Linvill for technical assistance.