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The promise of a fast-cooked healthy meal: common genomic regions for cooking time and iron and zinc content in beans revealed by meta-analysis

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ABSTRACT

Short cooking time (CT) is a highly desired trait in common beans, and several quantitative trait loci (QTL) were identified for bean grain quality traits (GQT), including CT. However, although several correlations were reported among these traits, these genomic regions were limitedly integrated and characterized. In this study, we collected 245 QTL for bean GQT from 12 mapping studies, of which 55 were for CT. A consensus map of the QTL was built based on the *Phaseolus vulgaris* G19833 genome v2.0, using QTL flanking positions when only these are available or physical positions corrected with study-reported linkage disequilibrium (LD) extent or a standard LD of 300 kbp to define QTL end and start positions. Then, the genes with the genomic regions concerning CT were prioritized through a series of in-silico analyses, such as keyword searches and protein-protein interaction networks. We resolved the map to 106 genomic regions, of which 27 were associated with multiple traits. Seventeen multi-trait (MTGR) and 17 single-trait genomic regions (STGR) involved QTL for CT. Nine CT-related MTGRs involved either or both Fe and Zn content, indicating a possibility of combining these traits in new varieties. These results generally agree with a previous meta-analysis conducted for Fe and Zinc QTL. We prioritized 566 that showed a significant PPI at the default confidence level of 0.4 (PPI enrichment p-value<1.0e-16), of which 122 were coexpressed, 138 experimentally determined, 298 based on database annotation, and 480 based on text-mining. All four specific interactions involved 537 genes, and 54 presented all these. We conclude that CT is genomically linked to Fe and Zn content, controlled by multiple genes operating in a biological network. Therefore, multi-omics-assisted genomic selection methods would be the best approach for combining short CT with other essential GQT in new end-user accepted varieties to promote bean consumption and consumers health.

Keywords: Candidate gene prioritization, hard-to-cook phenomenon, in-silico genomics analyses, *Phaseolus vulgaris*, quantitative trait loci

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RÉSUMÉ

Un court temps de cuisson (CT) est une caractéristique très recherchée chez le haricot, et plusieurs loci de traits quantitatifs (QTL) ont été identifiés pour les caractéristiques de qualité du grain de haricot (GQT), y compris le CT. Cependant, bien que plusieurs corrélations aient été rapportées entre cest caractéristiques, ces régions génomiques ont été peu intégrées et caractérisées. Dans cette étude, nous avons rassemblé 245 QTL pour le GQT du haricot à partir de 12 études de cartographie genomique, dont 55 pour le CT. Une carte consensuelle des QTL a été construite sur la base du génome Phaseolus vulgaris G19833 v2.0, en utilisant les positions des flancs des QTL lorsque seules celles-ci sont disponibles ou les positions physiques corrigées avec l'étendue du déséquilibre de liaison (LD) rapporté par l'étude ou un LD standard de 300 kbp pour définir les positions de début et de fin des QTL. Ensuite, les gènes localisee dans les régions génomiques concernent le CT ont été classés par ordre de priorité à l'aide d'une série d'analyses in-silico, telles que des recherches par mots-clés et des réseaux d'interactions protéine-protéine. Nous avons résolu la carte à 106 régions génomiques, dont 27 sont associées à des traits multiples. Dix-sept régions génomiques à caractères multiples (MTGR) et 17 régions génomiques à caractère unique (STGR) impliquent des QTL pour le TC. Neuf MTGR liés à CT impliquaient soit la teneur en Fe et en Zn, soit les deux, ce qui indique une possibilité de combiner ces caractères dans les nouvelles variétés. Ces résultats sont en grande partie en accord avec une méta-analyse précédente menée pour les QTL de Fe et de Zinc. Apres leclassement par ordre de priorité, 566 genes presentant des PPI significatives suivant le niveau de confiance par défaut de 0,4 (valeur d'enrichissement duPPI p<1,0e-16), dont 122 étaient coexprimées, 138 déterminées expérimentalement, 298 basées sur l'annotation de bases de données et 480 basées sur l'exploration de texte. Ces quatre interactions combinées ont impliqué 537 gènes, dont 54 presentant toutes ces type de PPI. Nous concluons que le CT est génomiquement lié à la teneur en Fe et en Zn, contrôlée par de multiples gènes opérant dans un réseau biologique. Par conséquent, les méthodes de sélection génomique assistée par des techniques multi-omiques seraient la meilleure approche pour combiner le CT court avec d'autres GQT essentiels dans de nouvelles variétés acceptées par les utilisateurs finaux afin de promouvoir la consommation de haricots et la santé des consommateurs.

Mots clés: Analyses génomiques in-silico, hiérarchisation des gènes candidats, loci de traits quantitatifs, *Phaseolus vulgaris*, phénomène de difficulté de cuisson

INTRODUCTION

Common bean (*Phaseolus vulgaris*) is an essential food crop worldwide, especially in the developing world, where it serves as the primary source of protein and a crucial contributor to daily calorie uptake of resource-limited sections of the population. Also, beans provide nutrients such as complex carbohydrates, essential micronutrients, dietary fiber, vitamin B, and antioxidants to rural and urban populations (Castro-Guerrero *et al.,* 2016). Therefore, beans contribute significantly to maintaining public health in regions where public hospital services are rudimentary and populations usually cannot afford to use these facilities (Hayat *et al.,* 2014).

Bioavailability and quality of bean nutrients and their consumption through bean meals are highly affected by the long cooking time (CT) that characterizes most bean varieties either through denaturation or consumer deterrence owing to high woodfuel cost (Feitosa *et al.,* 2018; Wiesinger *et* *al.,* 2018, 2020). These disadvantages of the hard-to-cook phenomenon in beans have spurred several breeding programs targeting the development of fast-cooking varieties. These breeding programs also improve other grain quality traits (GQT) that are essential to consumers and processors along CT, especially iron (Fe) and zinc (Zn) content and bioavailability and seed coat postharvest darkening (PHD) (Haman *et al.,* 2020; Hummel *et al.,* 2020). Reports exist of various significant correlation levels among bean GQTs (Wiesinger *et al.,* 2018; Saradadevi *et al.,* 2021). We hypothesize that these levels of correlation are at least partly underlined by gene pleiotropy or close linkage, which eventually translates into overlaps of the different genomic regions or quantitative trait loci (QTL) controlling these traits.

In genomics-assisted breeding (GAB) of beans GQTs, several QTL have been detected in various genetic and environmental backgrounds for CT and water absorption capacity (WAC) (Cichy *et al.,* 2015; Berry *et al.,* 2020; Bassett *et al.,* 2021; de Almeida *et al.,* 2021; Delfini *et al.,* 2021; Diaz *et al.,* 2021; Sadohara *et al.,* 2022), Fe and Zn content and bioavailability (Caproni *et al.,* 2020; Diaz *et al.,* 2020; Gunja a *et al.,* 2021; Katuuramu *et al.,* 2018; Nazir *et al.,* 2022), and SCPHD (Bassett *et al.,* 2021; Sadohara *et al.,* 2022). Also, the biochemical pathways controlling these traits were extensively studied (Astudillo-Reyes *et al.,* 2015; Izquierdo *et al.,* 2018; Wiesinger *et al.,* 2021; Toili *et al.,* 2022), prompting their intergration in beans breeding programs targeting one or several of these traits. However, this information (data) has not yet been integrated, limiting its usefulness in GAB, especially in breeding programs targeting simultaneous improvement and combining several GQTs in superior bean varieties. Integration of QTL data can generally be done through meta-analysis using statistical software such as BioMercator (Sosnowski *et al.,* 2012) when QTL flanking marker information is sufficient and common across studies, as was done for bean grain Fe and Zn content (Izquierdo *et al.,* 2018). In the cases of diverging flanking markers or insufficient information, a workable approach is aligning the QTL regions to a reference genome using various criteria (Wisser *et al.,* 2011). Therefore, in this study we meta-analyzed QTL for cooking time (CT), iron (Fe) and zinc (Zn) content, and seed coat postharvest darking, and prioritized candidate genes within the CT-related genomic regions using in-silico approaches.

METHODOLOGY

A flowchart of the methodology is presented in Figure 1. Briefly, we collected 245 QTL for bean GQT from 12 mapping studies, of which 55 were for CT, 72 for WAC, 54 for Fe content (5 for Fe bioavailability), 58 for Zn content, and 6 for PHD. We built a consensus map anchoring the physical positions of the QTL on the *Phaseolus vulgaris* G19833 genome v2.0 available on the Legumes Information System (LIS, https://legacy .legumeinfo.org/ genomes/gbrowse/phavu. G19833.gnm2). For that, we used QTL flanking positions when only these are available or physical positions corrected with study-reported linkage disequilibrium (LD) extent or a standard LD of 300 kbp upstream and downstream of the QTL physical position to define QTL end and start positions. We defined single-trait (STGR) and multi-trait genomic regions (MTGR) based on the overlapping of QTLs from different studies for different traits in the same regions. Genes within the CT-related MTGRs and STGRs were retrieved as pre-candidate genes (pCG), and their gene features, including gene descriptions, gene ontology, and protein domain information, and protein sequences, were retrieved through the BioMart tools of EnsemblPlants (https:// plants. ensembl.org/biomart/). The gene features were used to select pre-CT-related CG (pCT-CG) using the cell wall biosynthesis and Fe and Zinc accumulation-related keywords identified from a literature review (Astudillo-Reyes *et al.,* 2015; Izquierdo *et al.,* 2018; Wiesinger *et al.,* 2021; Toili *et al.,* 2022). The protein sequences of these pCT-CGs were used to identify the biologically connected genes through a protein-protein interaction network performed on STRING 11.5 (Szklarczyk *et al.,* 2019), which were selected as final CGs.

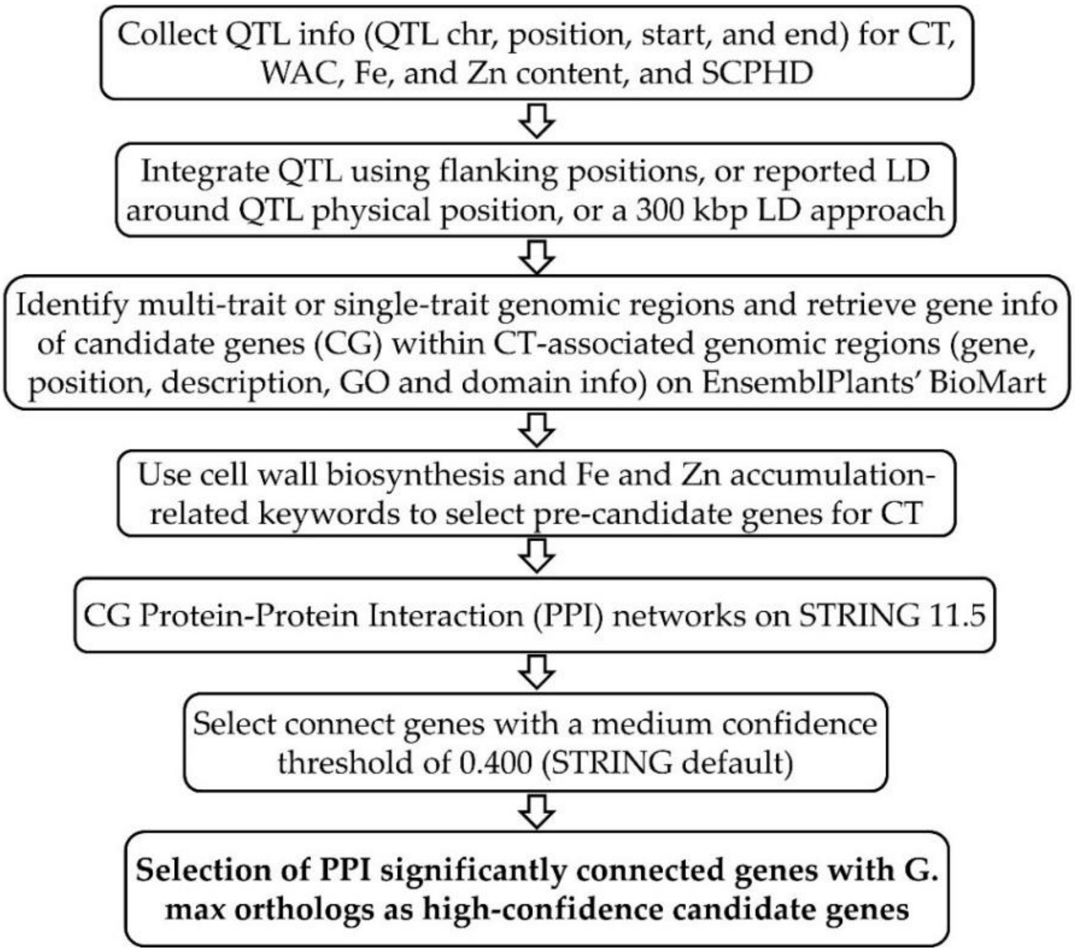

Figure 1. Flowchart of the methods used to integrate the QTLs and prioritize candidate genes for cooking time-related MTGRs and STGRs

RESULTS AND DISCUSSION

Genomic regions for combining short cooking time and high iron and zinc contents. Bean GQT, such as cooking time and color, are highly desired by consumers and processors, which determines the crop's commercial value. However, regarding health, the mineral content of the grain, mainly Fe and Zn, is highly valued. Therefore, breeding efforts are underway to combine these traits in new bean varieties worldwide (Mukankusi *et al.*, 2019). This research generates foundation information for using genomics-assisted breeding (GAB) to combine cooking time with nutrient (Fe and Zn) content in bean varieties. The QTL meta-analysis resolved the physical map to 106 genomic regions (Figure 2), of which 28 are associated with multiple traits. Seventeen multi-trait genomic regions (MTGR) and 18 single-trait genomic regions (STGR) involved QTL for CT.

Furthermore, nine CT-related MTGRs involved either or both Fe and Zn content, indicating a possibility of combining these traits in new varieties. These colocalizations between QTL for cooking time and iron and zinc content substantiate the correlations reported between these traits (Wiesinger *et al.*, 2018; Saradadevi *et al.*, 2021). Multivariate GWAS analyses would allow deciphering the true nature of these colocalizations, whether gene pleiotropy or close linkage controls these trait correlations (Schulthess *et al.,* 2017; Chebib and Guillaume, 2019). It is essential to characterize these genomic regions further to pinpoint the true nature of the colocalization, as this will guide future breeding ventures aiming at combining short cooking time with high bioavailable iron and zinc content (Sibov *et al.,* 2003; Schulthess *et al.,* 2017). There was no colocalization between cooking time and seed coat PHD, despite the two traits sharing similar molecular mechanisms under the phenylpropanoids biosynthetic pathway (Wiesinger *et al.,* 2021). However, this could be due to the extent of LD assumed to define QTL flanking positions. For common bean, reported LD decays vary largely from around 50 kbp (Wen *et al.,* 2019) to 4 Mbp (Gunja a *et al.,* 2021).

Comparing our results with another meta-analysis focusing on Fe and Zn content in common beans (Izquierdo *et al.,* 2018) revealed several convergences of the genomic regions controlling these two traits (Figure 3). However, some of the meta-QTL that Izquierdo *et al*. (2018) identified were not present in our map because their study covered some QTL for Fe and Zn that we did not include in our study because of the unavailability of marker sequence and physical positions. Given the high number of genomic regions involved in the control of bean cooking time and iron and zinc content, it is clear that traditional marker-assisted selection approaches are not appropriate for improving these traits either individually or in combination (Izquierdo *et al.,* 2018). The plethora of genes, biochemical pathways, and molecules determining bean cooking time and iron and zinc content, and bioavailability supports the need for multivariate genomic selection approaches to combine these traits in novel varieties (Astudillo-Reyes *et al.,* 2015; Izquierdo *et al.,* 2018; Wiesinger *et al.,* 2021; Toili *et al.,* 2022).

The genes with the CT-related genomic regions evidenced significant interactions. We identified 4191 genes within the CT-related MTGR and STGR, which were further reduced to 1092 genes using the keyword-based approach. Of these, 566 showed evidence of protein-protein interaction (PPI) at the default confidence level of 0.4 (PPI enrichment p-value<1.0e-^{16}), of which 122 for co-expression (p-value=2.77e-^{05}), 138 experimentally determined (p-value=0.00017), 298 based on database annotation (p-value=3.87e-^{09}), 480 based on text-mining (p-value=1.78e-^{08}), with all four specific interactions involving 537 genes (p-value<1.0e-^{16}) (Figure 4). The highly significant interaction levels among the proteins of the CGs with CT-related genomic regions that also involve other GQT, such as Fe and Zn content, indicate that these traits are genomically and biologically connected, controlled by multiple genes operating in a biological network, which is characteristics of genes involved in large biosynthetic pathways. These findings underscore the necessity of using multivariate and multi-omics-assisted genomic selection approaches for combining short CT with other essential GQT in new end-user accepted varieties.

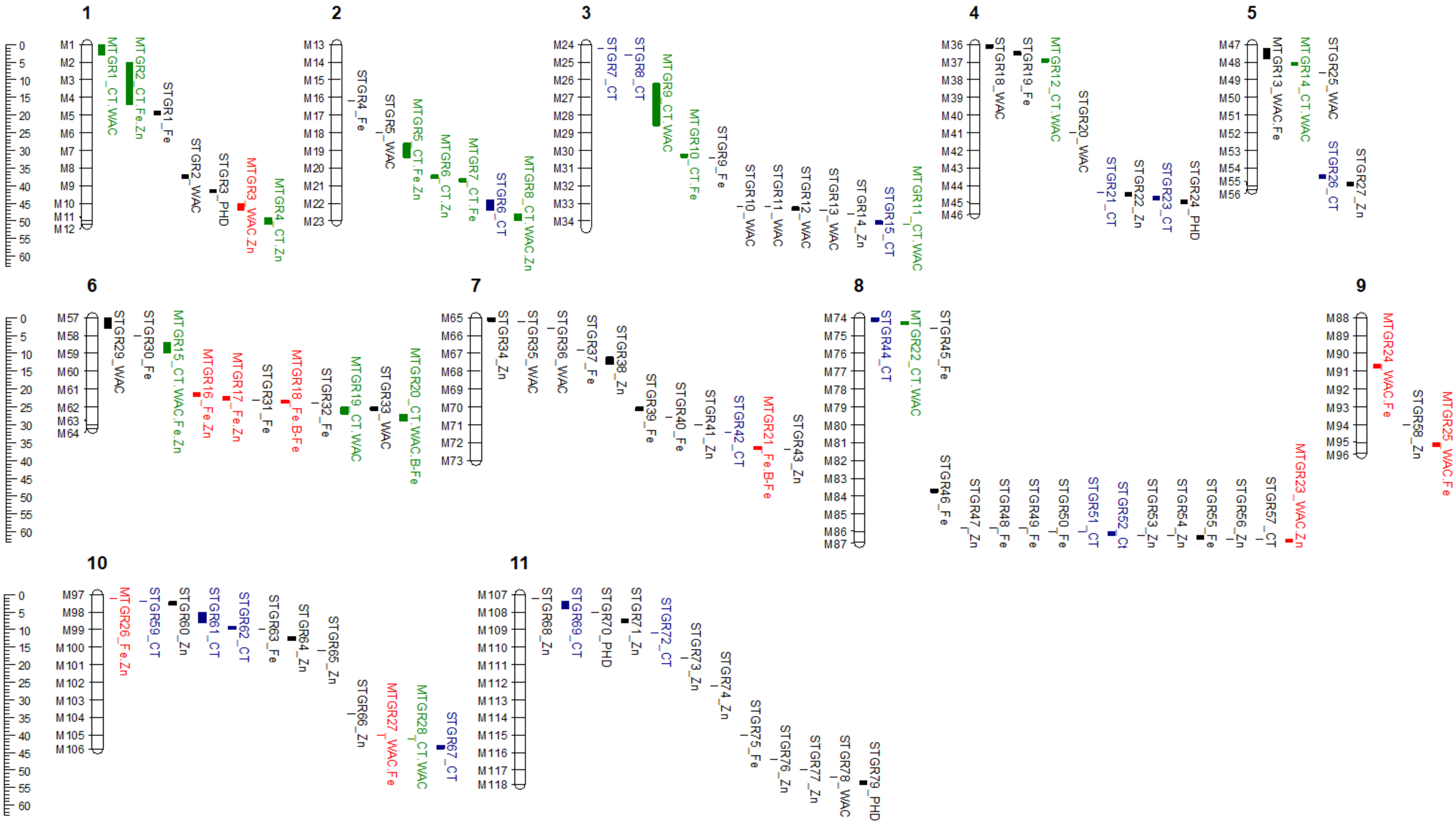

Figure 2. Meta-map of Multi-trait (MTGR) and Single-trait (STGR) genomic regions controling grain quality traits (GQT). In green: MTGR combining cooking time (CT) and other GQT, in blue: STGR unique to CT, and in red: MTGR involving other traits

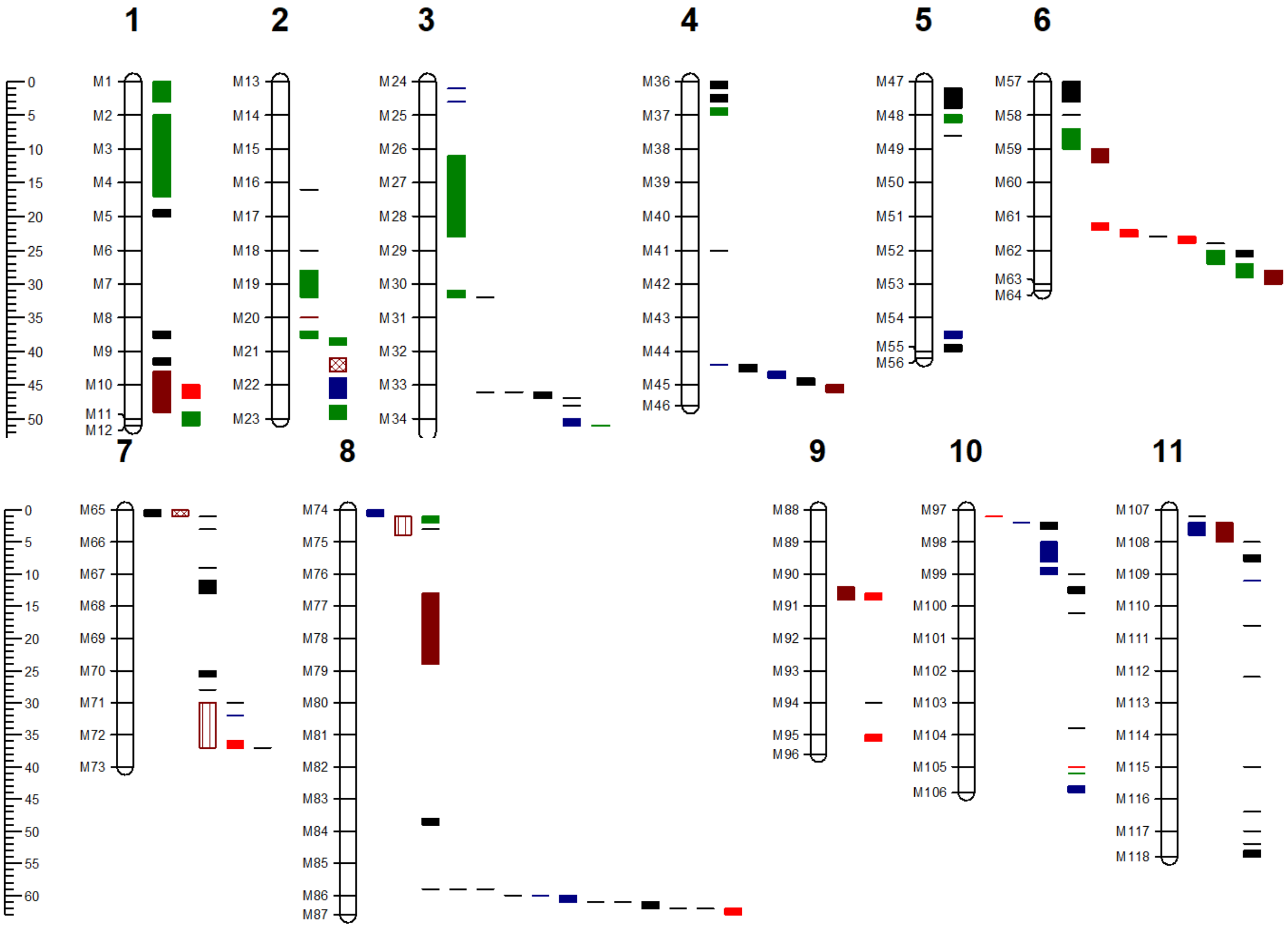

Figure 3. Meta-map positioning the MQTL for iron and zinc content (brown [in full brown: MQTL for Fe and Zinc; in brown with vertical strips: MQTLs for Fe, and in brown with crossed strips: MQTL for Zn]) from (Izquierdo *et al.*, 2018) relative to the MTGR and STGR GQT identified in this study.

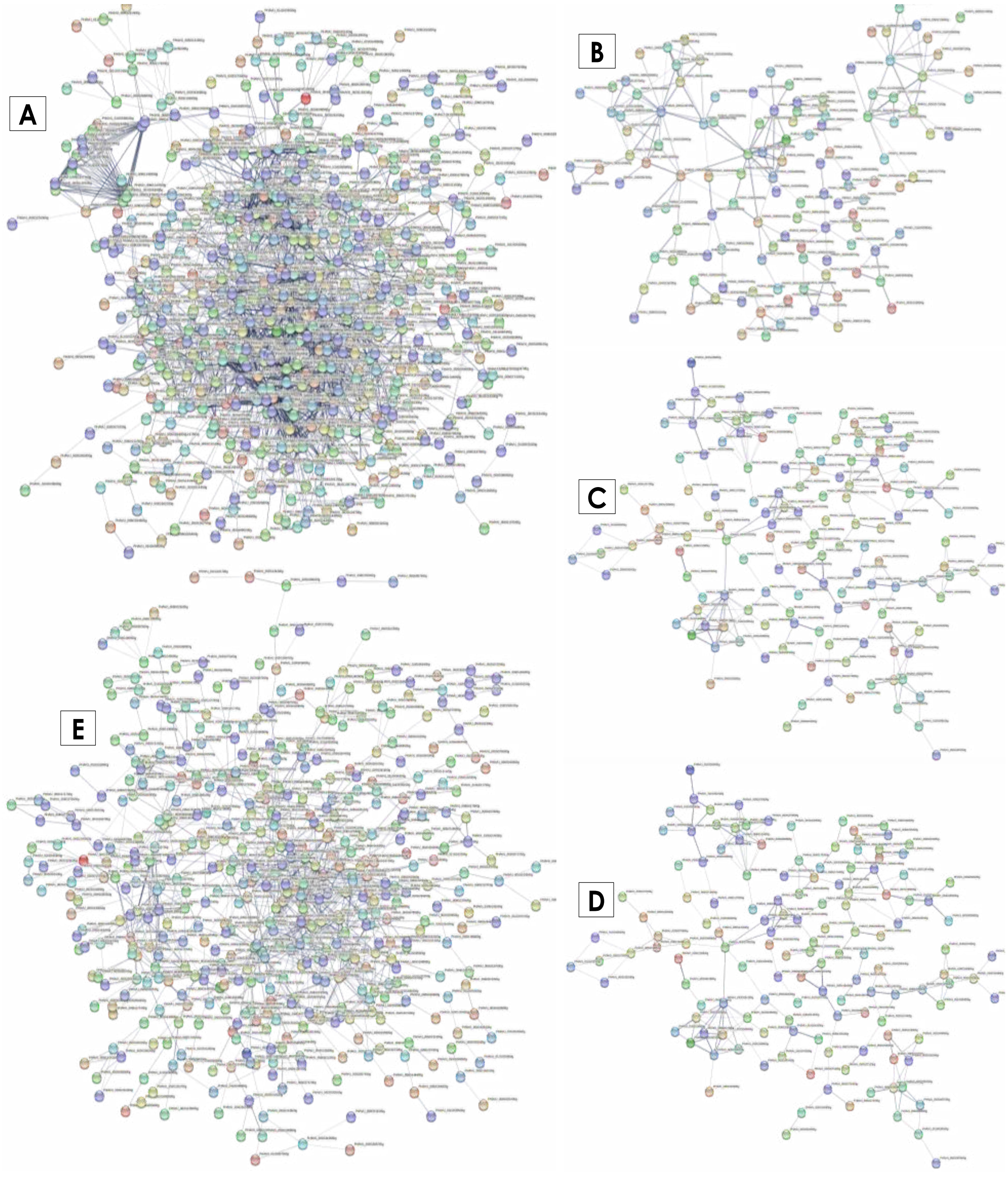

Figure 4. Protein-Protein interaction networks among the 566 candidate genes (excluding non-connected genes). A: Overall interaction, B: Coexpression, C: Database-determined, D: Text-mining, E: Experimentally-determined.

CONCLUSION

We conducted the first meta-analysis study investigating the genomic and gene-based relationship between cooking time and other GQT, such as cooking time, mineral (Fe and Zn) content, and seed coat PHD. Several colocalizations between CT and both or either Fe and Zn were revealed, and genes under these genomic regions shared extensive protein-protein interactions. There were no common genomic regions between CT and PHD, although these two types of QTL were in the vicinity of one another. Based on these findings, we recommend using multivariate and multi-omics-assisted genomic selection approaches to combine these GQT in novel end-users accepted bean varieties to promote consumption and consumers' health.

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STATEMENT OF NO CONFLICT OF INTEREST

The authors declare that there are no competing interests in this publication.

REFERENCES

Astudillo-Reyes, C., Fernandez, A. C. and Cichy, K. A. 2015. Transcriptome characterization of developing bean (*Phaseolus vulgaris* L.) pods from two genotypes with contrasting seed zinc concentrations. *PLoS ONE* 10 (9): 1–18. https://doi.org/10.1371/journal.pone.0137157

Bassett, A., Katuuramu, D. N., Song, Q. and Cichy, K. 2021. QTL mapping of seed quality traits including cooking time, flavor, and texture in a Yellow Dry Bean (*Phaseolus vulgaris* L.) Population. *Frontiers in Plant Science* 12: 1–16. https://doi.org/10.3389/fpls.2021.670284

Berry, M., Izquierdo, P., Jeffery, H., Shaw, S., Nchimbi-Msolla, S. and Cichy, K. 2020. QTL analysis of cooking time and quality traits in dry bean (Phaseolus vulgaris L.). *Theoretical and Applied Genetics* 133(7): 2291–2305. https://doi.org/10.1007/s00122-020-03598-w

Caproni, L., Raggi, L., Talsma, E. F., Wenzl, P. and Negri, V. 2020. European landrace diversity for common bean biofortification: a genome-wide association study. *Scientific Reports*10 (1): 3–7. https://doi.org/10.1038/s41598-020-76417-3

Castro-Guerrero, N. A., Isidra-Arellano, M. C., Mendoza-Cozatl, D. G. and Valdés-López, O. 2016. Common Bean: A legume model on the rise for unraveling responses and adaptations to iron, zinc, and phosphate deficiencies. *Frontiers in Plant Science* 7: 391–417. https://doi.org/10.3389/fpls.2016.00600

Chebib, J. and Guillaume, F. 2019. Pleiotropy or linkage? Their relative contributions to the genetic correlation of quantitative traits and detection by multi-trait GWA studies. BioRxiv. https://doi.org/10.1101/656413

Cichy, K. A., Wiesinger, J. A. and Mendoza, F. A. 2015. Genetic diversity and genome-wide association analysis of cooking time in dry bean (*Phaseolus vulgaris* L.). *Theoretical and Applied Genetics* 128 (8): 1555–1567. https://doi.org/10.1007/s00122-015-2531-z

de Almeida, C. P., Santos, I. L., de Carvalho Paulino, J. F., Barbosa, C. C. F., Pereira, C. C. A., Carvalho, C. R. L., de Moraes Cunha Gonçalves, G., Song, Q., Carbonell, S. A. M., Chiorato, A. F. and Benchimol-Reis, L. L. 2021. Genome-wide association mapping reveals new loci associated with light-colored seed coat at harvest and slow darkening in carioca beans. *BMC Plant Biology* 21(1): 1–15. https://doi.org/10.1186/s12870-021-03122-2

Delfini, J., Moda-Cirino, V., dos Santos Neto, J., Zeffa, D. M., Nogueira, A. F., Ribeiro, L. A. B., Ruas, P. M., Gepts, P. and Gonçalves, L. S. A. 2021. Genome-wide association study identifies genomic regions for important morpho-agronomic traits in Mesoamerican Common Bean. *Frontiers in Plant Science* 12: 1–14. https://doi.org/10.3389/fpls.2021.748829

Diaz, S., Ariza-Suarez, D., Izquierdo, P., Lobaton, J. D., de la Hoz, J. F., Acevedo, F., Duitama, J., Guerrero, A. F., Cajiao, C., Mayor, V., Beebe, S. E. and Raatz, B. 2020. Genetic mapping for agronomic traits in a MAGIC population of common bean (*Phaseolus vulgaris* L.) under drought conditions. *BMC Genomics* 21 (1): 1–21.https://doi.org/10.1186/s12864-020-07213-6

Diaz, S., Ariza-Suarez, D., Ramdeen, R., Aparicio, J., Arunachalam, N., Hernandez, C., Diaz, H., Ruiz, H., Piepho, H. P. and Raatz, B. 2021. Genetic architecture and genomic prediction of cooking time in Common Bean (*Phaseolus vulgaris* L.). *Frontiers in Plant Science* 11(February). https://doi.org/10.3389/fpls.2020.622213

Feitosa, S., Greiner, R., Meinhardt, A. K., Müller, A., Almeida, D. T. and Posten, C. 2018. Effect of traditional household processes on iron, zinc and copper bioaccessibility in black bean (*Phaseolus vulgaris* L.). *Foods* 7(8): 1–12. https://doi.org/10.3390/foods7080123

Gunja a, J., Carovi -Stanko, K., Lazarevi , B., Vidak, M., Petek, M., Liber, Z. and Šatovi, Z. 2021. Genome-wide association studies of mineral content in Common Bean. *Frontiers in Plant Science* 12:1–12. https://doi.org/10.3389/fpls.2021.636484

Haman, Y., Abatchoua, A. and Tchiagam, N. 2020. Genetic analysis of cooking time in five Common Bean (*Phaseolus vulgaris* L.) cultivars grown in Cameroon. *EAS Journal of Biotechnology and Genetics* 2 (4):58-66.

Hayat, I., Ahmad, A., Masud, T., Ahmed, A. and Bashir, S. 2014. Nutritional and health perspectives of beans (*Phaseolus vulgaris* L.): An overview. *Critical Reviews in Food Science and Nutrition* 54 (5): 580–592. https://doi.org/10.1080/10408398.2011.596639

Hummel, M., Talsma, E. F., Taleon, V., Londoño, L., Brychkova, G., Gallego, S., Raatz, B. and Spillane, C. 2020. Iron, zinc and phytic acid retention of biofortified, low phytic acid, and conventional bean varieties when preparing common household recipes. *Nutrient*s 12(3): 658-. https://doi.org/10.3390/nu12030658

Izquierdo, P., Astudillo, C., Blair, M. W., Iqbal, A. M., Raatz, B. and Cichy, K. A. 2018. Meta-QTL analysis of seed iron and zinc concentration and content in common bean (*Phaseolus vulgaris* L.). *Theoretical and Applied Genetics* 131(8): 1645–1658. https://doi.org/10.1007/s00122-018-3104-8

Katuuramu, D. N., Hart, J. P., Porch, T. G., Grusak, M. A., Glahn, R. P. and Cichy, K. A. 2018. Genome-wide association analysis of nutritional composition-related traits and iron bioavailability in cooked dry beans (*Phaseolus vulgaris* L .). *Molecular Breeding* 38: 1-18.

Mukankusi, C., Raatz, B., Nkalubo, S., Berhanu, F., Binagwa, P., Kilango, M., Williams, M., Enid, K., Chirwa, R. and Beebe, S. 2019. Genomics, genetics and breeding of common bean in Africa: A review of tropical legume project. *Plant Breeding* 138(4): 401–414. https://doi.org/10.1111/pbr.12573

Nazir, M., Mahajan, R., Mansoor, S., Rasool, S., Mir, R. A., Singh, R., Thakral, V., Kumar, V., Sofi, P. A., El-Serehy, H. A., Hefft, D. I. and Zargar, S. M. 2022. Identification of QTLs/candidate genes for seed mineral contents in Common Bean (*Phaseolus vulgaris* L.) Through Genotyping-by-Sequencing. *Frontiers in Genetics* 13: 1–15. https://doi.org/10.3389/fgene.2022.750814

Sadohara, R., Izquierdo, P., Couto Alves, F., Porch, T., Beaver, J., Urrea, C. A. and Cichy, K. 2022. The *Phaseolus vulgaris* L. yellow bean collection: genetic diversity and characterization for cooking time. *Genetic Resources and Crop Evolution* 69 (4): 1627–1648. https://doi.org/10.1007/s10722-021-01323-0

Saradadevi, R., Mukankusi, C., Li, L., Amongi, W., Mbiu, J. P., Raatz, B., Ariza, D., Beebe, S., Varshney, R. K., Huttner, E., Kinghorn, B., Banks, R., Rubyogo, J. C., Siddique, K. H. M. and Cowling, W. A. 2021. Multivariate genomic analysis and optimal contributions selection predicts high genetic gains in cooking time, iron, zinc, and grain yield in common beans in East Africa. *Plant Genome* 14(3): 1–16. https://doi.org/10.1002/tpg2.20156

Schulthess, A. W., Reif, J. C., Ling, J., Plieske, J., Kollers, S., Ebmeyer, E., Korzun, V., Argillier, O., Stiewe, G., Ganal, M. W., Röder, M. S. and Jiang, Y. 2017. The roles of pleiotropy and close linkage as revealed by association mapping of yield and correlated traits of wheat (*Triticum aestivum* L.). *Journal of Experimental Botany* 68 (15): 4089–4101. https://doi.org/10.1093/jxb/erx214

Sibov, S. T., Souza Junior, C. L. de, Garcia, A.

A. F., Silva, A. R., Garcia, A. F., Mangolin, C. A., Benchimol, L. L. and De Souza, A. P. 2003. Molecular mapping in tropical maize (*Zea mays* L.) using microsatellite markers. 2. Quantitative trait loci (QTL) for grain yield, plant height, ear height and grain moisture. *Hereditas* 139(2): 107–115. https://doi.org/10.1111/j.1601-5223.2003.01667.x

Sosnowski, O., Charcosset, A. and Joets, J. 2012. Biomercator V3: An upgrade of genetic map compilation and quantitative trait loci meta-analysis algorithms. *Bioinformatics* 28(15): 2082–2083. https://doi.org/10.1093/bioinformatics/bts313

Szklarczyk, D., Gable, A. L., Lyon, D., Junge, A., Wyder, S., Huerta-Cepas, J., Simonovic, M., Doncheva, N. T., Morris, J. H., Bork, P., Jensen, L. J. and Von Mering, C. 2019. STRING v11: Protein-protein association networks with increased coverage, supporting functional discovery in genome-wide experimental datasets. *Nucleic Acids Research* 47(D1): D607–D613. https://doi.org/10.1093/nar/gky1131

Toili, M. E. M., de Koning, R., Kiekens, R., Ndumba, N., Wahome, S., Anami, S., Githiri, S. M. and Angenon, G. 2022. Transcriptome-guided identification of pectin methyl-esterase-related enzymes and novel molecular processes effectuating the hard-to-cook defect in Common Bean (*Phaseolus vulgaris* L.). *Foods* 11(12): 1692. https://doi.org/10.3390/foods11121692

Wen, L., Chang, H. X., Brown, P. J., Domier, L. L. and Hartman, G. L. 2019. Genome-wide association and genomic prediction identifies soybean cyst nematode resistance in common bean including a syntenic region to soybean Rhg1 locus. *Horticulture Research* 6(1):. https://doi.org/10.1038/s41438-018-0085-3

Wiesinger, J. A., Cichy, K. A., Hooper, S. D., Hart, J. J. and Glahn, R. P. 2020. Processing white or yellow dry beans (*Phaseolus vulgaris* L.) into a heat treated flour enhances the iron bioavailability of bean-based pastas. *Journal of Functional Foods* 71: 104018. https://doi.org/10.1016/j.jff.2020.104018

Wiesinger, J. A., Cichy, K. A., Tako, E. and Glahn, R. P. 2018. The fast cooking and enhanced iron bioavailability properties of the manteca yellow bean (*Phaseolus vulgaris* L.). *Nutrients* 10(11). https://doi.org/10.3390/nu10111609

Wiesinger, J. A., Osorno, J. M., McClean, P. E., Hart, J. J. and Glahn, R. P. 2021. Faster cooking times and improved iron bioavailability are associated with the down regulation of procyanidin synthesis in slow-darkening pinto beans (*Phaseolus vulgaris* L.). *Journal of Functional Foods* 82: 104444. https://doi.org/10.1016/j.jff.2021.104444

Wisser, R. J., Kolkman, J. M., Patzoldt, M. E., Holland, J. B., Yu, J., Krakowsky, M., Nelson, R. J. and Balint-Kurti, P. J. 2011. Multivariate analysis of maize disease resistances suggests a pleiotropic genetic basis and implicates a GST gene. *Proceedings of the National Academy of Sciences* 108 (18): 7339–7344. https://doi.org/10.1073/pnas.1011739108