## GENOMICS OF MULTIPLE-INSECT RESISTANCE IN MAIZE

Abstract: Maize, a food security and multipurpose crop, is attacked on all parts of the plant, from the fields to the storage facilities, by combinations of a plethora of concurrently or subsequently occurring insect pests. However, most studies conducted to understand maize defense mechanisms, targeting to incorporate resistance in new varieties, focused on single insect attacks. Nevertheless, phenotypic correlations and colocalizations of quantitative trait loci (QTL) for maize resistance to stem borers (SB), the fall armyworm (FAW), and storage pests (SP) on one hand, and overlaps with cell wall constituents (CWC) and benzoxazinoids (BXs) QTL, on the other hand, suggests the existence of multiple-insect resistance (MIR) resistance mechanisms. Therefore, the goal of this research was to investigate the feasibility of MIR in maize through the application of genomic-aided breeding (GAB) technologies for mapping and selection by establishing the genetic architecture and genomic predictability of maize resistance to multiple insect pests. A tissue-specific meta-analysis of QTL for resistance to SB, FAW, SP, CWC, and BXs revealed 42 stem (SIR), 24 leaf (LIR), and 20 kernel (KIR) insect resistance meta-QTL (MQTL) which involved each at least one CWC or BXs, suggesting a defensive role played by these biochemicals in resistance. Seven genomic regions containing overlaps among resistance MQTL in different tissues of which, 3 involved KIR, SIR, and LIR resistance categories, which is a promise for combining resistance to both damage and aflatoxin contaminations in maize varieties. The commonality of insect-resistance genomic regions (IRGRs) across genetic and geographic background and the common genomic regions identified for damage resistance on maize plants and grains revealed in this study motivated further investigations of insect resistance genomic regions in local environments for locally occurring insect pests such as FAW and MW. An association mapping panel (AMP) composed of 341 genetically and geographically Africa-adapted lines was planted in Kasese (2017B) and Namulonge (2018A and B) in an augmented design and genotyped with 34509 diversity array technology (DArT) single-nucleotide polymorphism (SNP) markers. Removal of duplicate SNP rendered 28919 markers (DRSNPS) and then, linkage disequilibrium (LD)-based pruning reduced the genotypes to 3124 SNPs (LDSNPs). The 341 lines of the AMP were phenotyped for FAW damage resistance, and subsequently, grains 126 lines bulked across environments were evaluated for MW resistance traits such as grain weight loss (GWL), flour production (FP), adult progeny emergence (AP), number of affected kernels (AK), and number of holes (NH). With the 3124 LDSNPs, a multi-locus GWAS mixed linear model incorporating both population structure as genotype-based principle components and kinship relatedness was conducted using the best linear unbiased predictors (BLUPs) generated from the MW resistance phenotypes and the across-environment FAW damage scores. This analysis revealed 62 quantitative trait nucleotides (QTNs) majorly located within or near the IRGRs, of which, six were associated with resistance to both FAW and MW suggesting a pleiotropic control. Besides, a set of In-silico interspecific comparative functional analyses discovered 79 candidate genes located around the QTNs, of which, 64 were functionally related to plant defense mechanisms, and only one candidate gene for each was identified for five of the six multiple-insect pest QTNs, corroborating the possible pleiotropic hypothesis. Further, an additional 107 candidate genes differentially expressed under various stress conditions and located within the IRGRs were identified through network-based inferences. These QTNs and genes could be incorporated in GAB, gene editing, and transgenic programs targeting either single or combined insect resistance. However, the

plethora of genomic regions involved in resistance implicated the necessity of incorporating genomic selection (GS) in insect resistance breeding programs. Therefore, a benchmarking of diverse Bayesian, mixed model, and Machine learning genomic prediction (GP) algorithms using both random (RBTS) and a pedigree-based training (PBTS) determination strategies was performed for FAW and MW resistance traits using the 28919 DRSNPs. The prediction accuracies (PA) were high for FAW (up to 86%), and MW resistance traits such as AP (up to 77%), GWL (up to 79%), and AP (up to 82%) even with small-sized TS (37% of the AMP), thus, pioneering the application of GS in breeding for resistance to insect pests. The GP models performed differently, with Bayesian models performing best on MW resistance traits while Machine learning and mixed model algorithms slightly outperformed Bayesian models on FAW resistance datasets. Also, the size of the TS and its genetic relationship with the BS was determinant, with both a positive and negative correlation of the TS size with PAs for RBTS and PBTS, respectively. Overall, this research sets the basis for scaling up the development of multiple-insect resistant maize varieties.

**Keywords:** Maize, Comparative and association mapping, candidate gene functional prioritization, Genomic prediction and selection, Combined resistance.

## **Publications:**

- Badji, A., Otim, M., Machida, L., ODONG, T. L., Kyamanywa, S., & Rubaihayo, P. (2018). Maize resistance to stem borers and storage pests: The need of new genetics and functional genomics approaches in future research. *African Journal of Rural Development*, 2(3), 467-479.
- Badji A, Otim M, Machida L, Odong T, Kwemoi DB, Okii D, Agbahoungba S, Mwila N, Kumi F, Ibanda A, Mugo S, Kyamanywa S and Rubaihayo P (2018) Maize Combined Insect Resistance Genomic Regions and Their Co-localization With Cell Wall Constituents Revealed by Tissue-Specific QTL Meta-Analyses. Front. Plant Sci. 9:895. doi: 10.3389/fpls.2018.00895.
- Badji, A.; Kwemoi, D.B.; Machida, L.; Okii, D.; Mwila, N.; Agbahoungba, S.; Kumi, F.; Ibanda, A.; Bararyenya, A.; Solemanegy, M.; Odong, T.; Wasswa, P.; Otim, M.; Asea, G.; Ochwo-Ssemakula, M.; Talwana, H.; Kyamanywa, S.; Rubaihayo, P. Genetic Basis of Maize Resistance to Multiple Insect Pests: Integrated Genome-Wide Comparative Mapping and Candidate Gene Prioritization. Genes 2020, 11, 689.doi: <u>https://doi.org/10.3390/genes11060689</u>.
- Badji, A.; Machida, L.; Kwemoi, D.B.; Kumi, F.; Okii, D.; Mwila, N.; Agbahoungba, S.; Ibanda, A.; Bararyenya, A.; Nghituwamhata, S.N.; Odong, T.; Wasswa, P.; Ochwo-Ssemakula, M.; Talwana, H.; Asea, G.; Otim, M.; Kyamanywa, S.; Rubaihayo, P. Factors Influencing Genomic Prediction Accuracies of Tropical Maize Resistance to Fall Armyworm and Weevils. Preprints 2020, 2020070336. doi: 10.20944/preprints202007.0336.v3.