

Connecting a prior-knowledge network with APSIM: Towards breeding by design

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Abstract: The transition from Breeding 3.0 to Breeding 4.0 requires moving beyond statistical associations toward knowledge-driven approaches. We present APSIM-PSoup, a framework that integrates qualitative prior-knowledge networks with crop models and has the potential to enable the design and testing of allele combinations *in silico* across different G×M contexts before field deployment. This proof-of-concept demonstrates how prior-knowledge networks can inform trait prediction, revealing G×E×M interactions and opening new avenues for breeding by design.

Keywords: Breeding 4.0, Prior-knowledge networks, PSoup, APSIM, G×E×M

1. INTRODUCTION

New approaches are required to enable the shift from Breeding 3.0, where genomic data are integrated into predictive pipelines, to Breeding 4.0 (Wallace et al. 2018), which aspires to design optimal allele combinations. Central to this transition is the use of knowledge networks, structured representations of causal gene, hormone, and regulatory interactions. These networks move us beyond statistical associations and enable breeding by design, where hypotheses about allele combinations and their emergent phenotypes can be tested *in silico* before field deployment. Current breeding pipelines that use prior knowledge networks often rely on heavy parameterization, which is crop-specific and difficult to generalize. To overcome this, we used PSoup, a package that encodes the topology of prior-knowledge networks without requiring parameters, and where the outcome is qualitative. By embedding these qualitative networks into the APSIM-sorghum model, we take a decisive step toward leveraging decades of molecular biology to inform trait prediction under diverse environments.

2. METHODOLOGY AND RESULTS

This APSIM-PSoup (<https://psoup-apsim.streamlit.app/>) framework involved two steps: (1) creating prior-knowledge networks using PSoup to test their predictive accuracy, and (2) integrating the prior-knowledge network into APSIM (Figure 1). As a case study, we constructed a regulatory network of shoot branching (ShootX; <https://shootx-psoup.streamlit.app/>), which improved upon network topologies given within the latest reviews for shoot branching (Beveridge et al. 2023; Dun et al. 2023). This network was developed from the analysis of 51 number of studies producing 81 number of unique perturbations. Perturbations via simulated mutants and treatments produced qualitative predictions (increased, decreased, unchanged branching) with 82.3% accuracy during training and 70.1% during testing. These qualitative outcomes of shoot branching for different genotypes were then mapped to APSIM tiller numbers through scaling rules that anchor qualitative classes to crop-scale ranges.

Our results from testing mutant scenarios across diverse environments and management schemes revealed clear G×E×M interactions. The framework was able to distinguish classes of mutant responses across different environment–management combinations, patterns that could not be captured by network simulations alone. A key challenge remains in translating qualitative predictions into quantitative scales, to ensure that the G×E×M interactions reflect realistic crop outcomes rather than remaining purely hypothetical. As PSoup encodes network topology rather than kinetic parameters, the few parameters required for mapping qualitative outcomes to quantitative values are likely to be readily estimated and more broadly applicable. Field data can later refine assumptions and provide parametrization where needed. Specifically, hypotheses can be tested from network behaviour to the field. For example, sucrose affects the protein MAX2, and using the shoot branching network it shows that increasing sucrose levels increase branching not only because of higher sucrose levels, but also by inhibiting strigolactone perception through MAX2 (Patil et al. 2022). Thus, a *max2* mutant produces more

branching than other mutants. However, when combined with management and environmental variation, new scenarios emerge such as where *max2* mutants behave differently from other mutants in the same pathway.

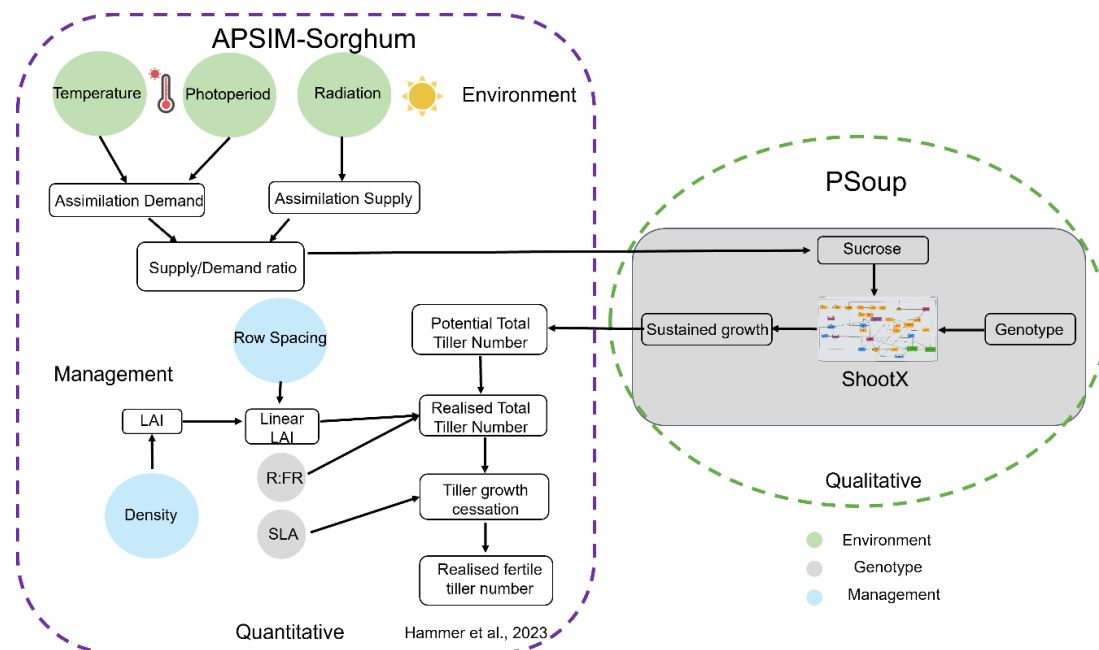


Figure 1. Conceptual representation of the APSIM-PSoup framework. Environmental drivers (temperature, photoperiod, radiation) define the supply–demand ratio, which is mapped to sucrose in the ShootX network. Genotypes are introduced into ShootX and simulated via the PSoup package, producing sustained growth as a qualitative outcome of shoot branching. Sustained growth is then converted into potential total tiller number through a scaling formula, providing a quantitative measure. Colour coding denotes inputs from environment (green), genotype (grey), and management (blue).

3. CONCLUSIONS

This work represents a proof-of-concept for creating and embedding qualitative prior knowledge into crop models to support the transition to Breeding 4.0. By systematically linking regulatory networks to quantitative outcomes, we open avenues for designing allele combinations guided by mechanistic understanding, testing hypotheses across environments before costly trials, and bridging the gap between QTLs and traits by having networks that are affected by QTLs and, in turn, shape complex traits such as yield (Cooper *et al.* 2021). Ultimately, this approach demonstrates how breeding by design can become reality: moving from statistical association to mechanistic integration, from model plants to crops, and from Breeding 3.0 to Breeding 4.0.

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