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# Effect of allelic forms of *GRFs* genes on the development of common wheat under different conditions of nitrogen supplementation



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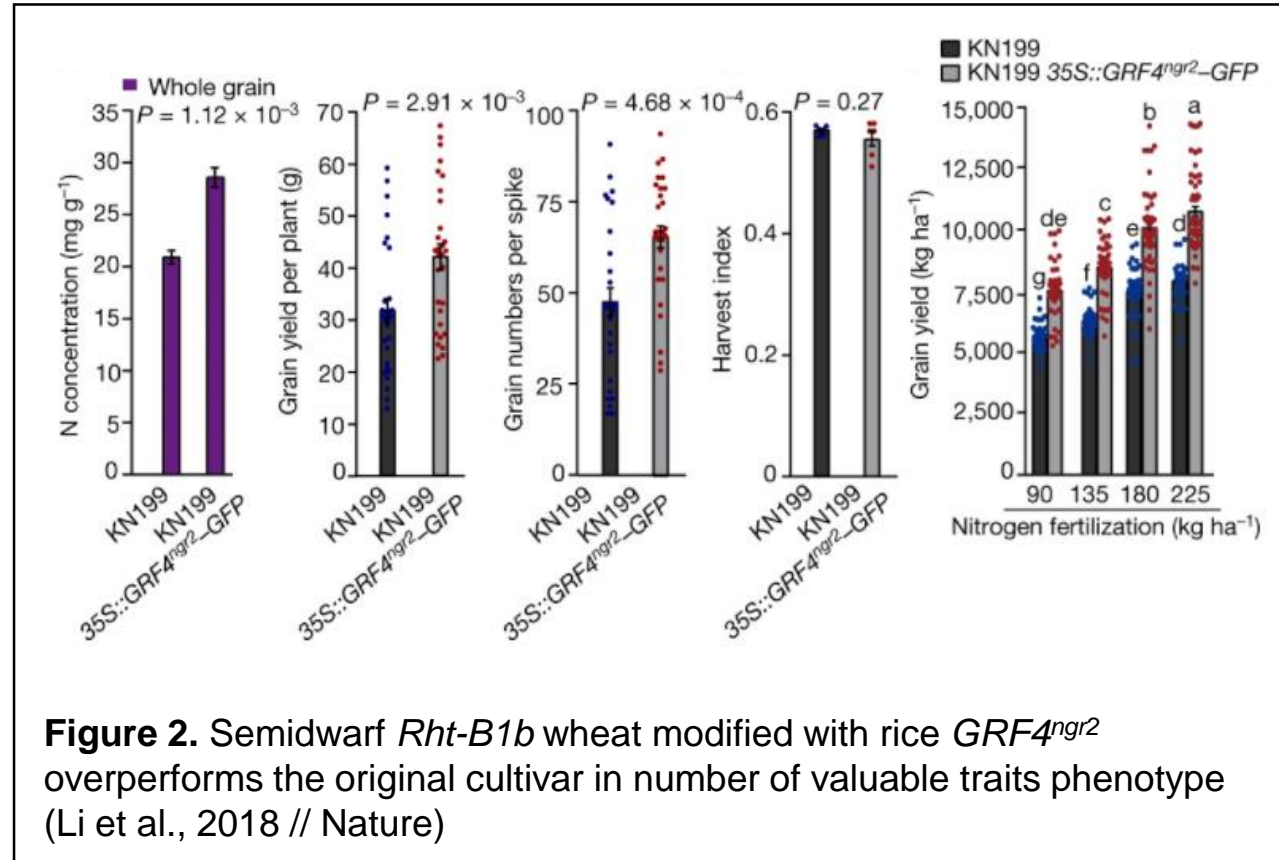
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# Introduction

- Nitrogen fertilizers increase crop yield, however the plants grow taller and became more susceptible to lodging.
- The reduced height (“dwarfing”) genes have been introduced into modern cereal cultivars to overcome the excessive plant growth in nitrogen-fertilized cultivation.
- Unfortunately, common dwarfing genes *Rht* reduce the response to nitrogen fertilizers and reduce the mass of 1000 grains.

- *GRFs* (Growth-regulating factors) are a family of genes that regulate plant growth and development, organ and tissue formation, and plant response to stress.

- *GRF4* improves agronomically important traits and increases the efficiency of nitrogen fertilization in rice and wheat (Figs.1,2).

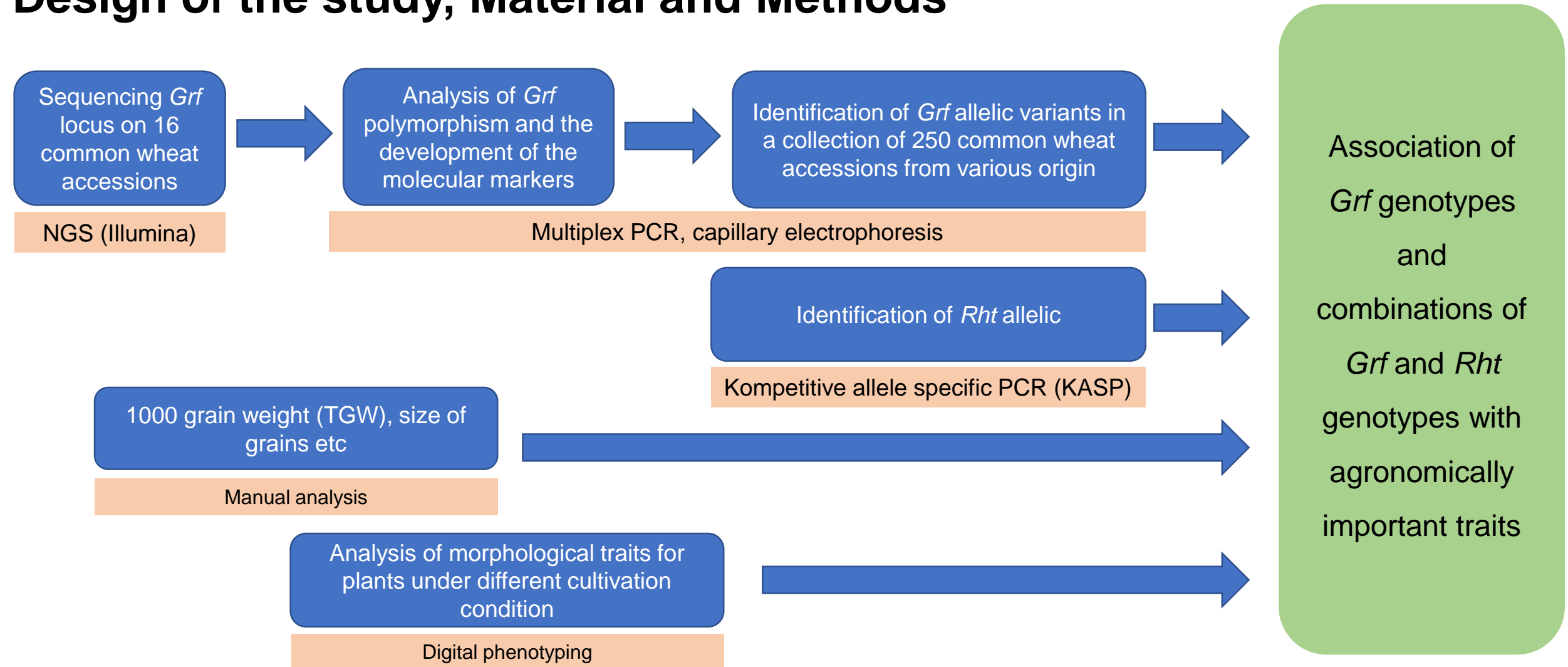


**Figure 1.** Panicle and grain is larger in rice plants with overexpressed *GRF4* than in control (Sun et al., 2016 // Journal of Integrative Plant Biology)

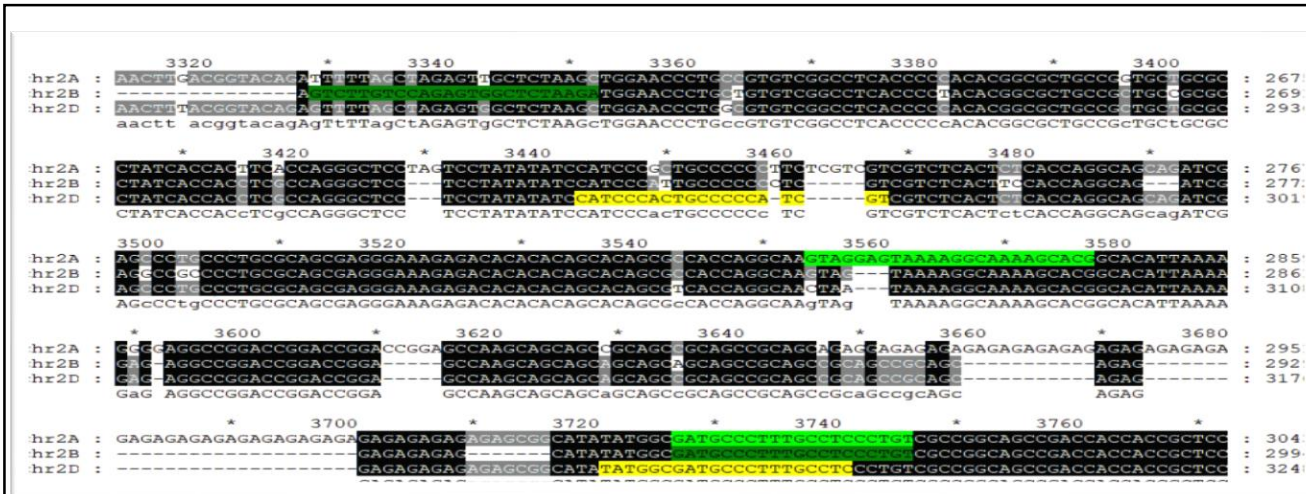
# Objectives

To investigate the effect of natural *GRF* alleles on the morphologic traits and yield of common wheat

## Design of the study, Material and Methods

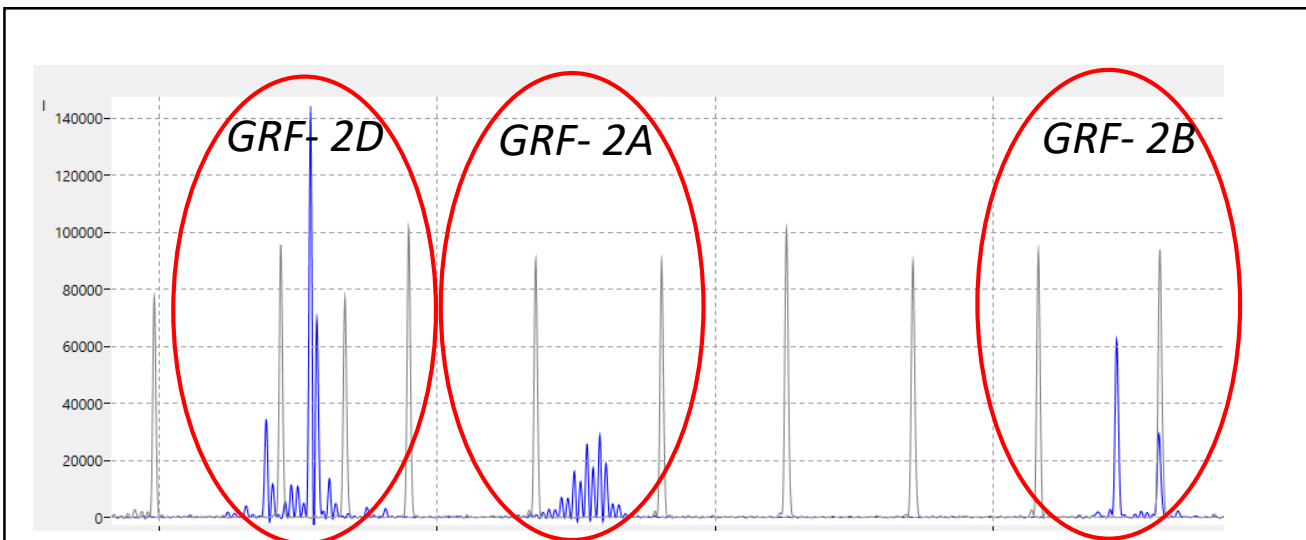


# Results

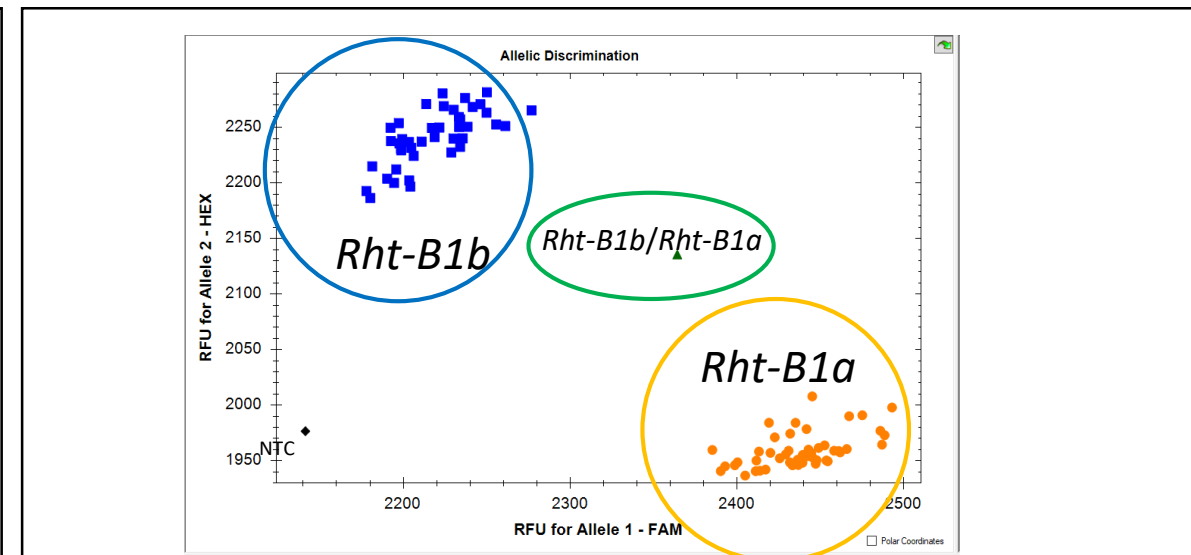


**Figure 3.** Multiple alignment of *Grf* alleles and primers developed for *GRF-2A*, *GRF-2B*, *GRF-2D* genotyping in common wheat.

- *Grf* locus have been sequenced in 16 common wheat cultivars and allelic variants have been revealed. *Grf-2A* turned out to be the most polymorphic.
- Molecular marker were developed for multiplex PCR genotyping (Fig. 3).
- A collection of 250 common wheat accessions was screened and large variety of alleles was found in *GRF-2A*, *GRF-2B*, *GRF-2D* genes (Fig. 4)
- Beside *Grf*, the wheat collection was genotyped for *Rht* (Fig. 5)
- An association of 1000-grain weight (TGW) with *GRF* – *Rht* combinations was observed.

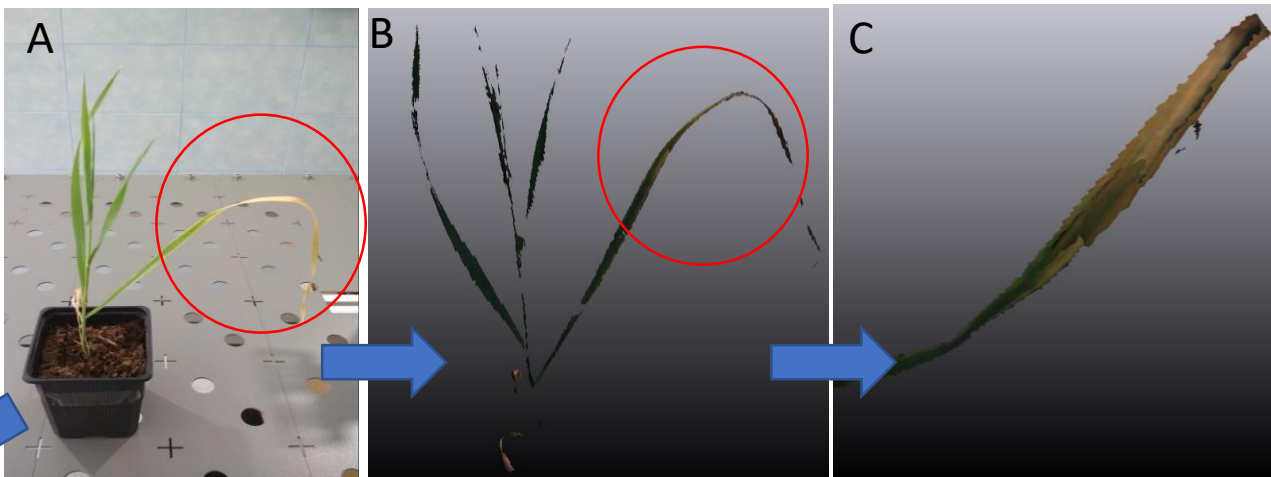


**Figure 4.** Fragment analysis of a wheat specimen.

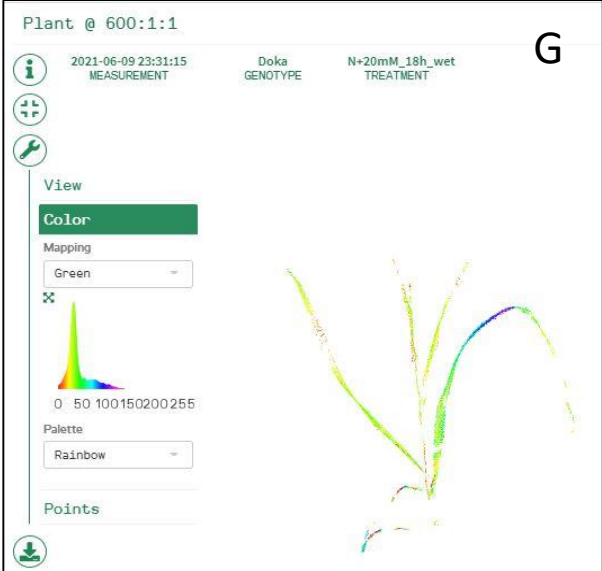
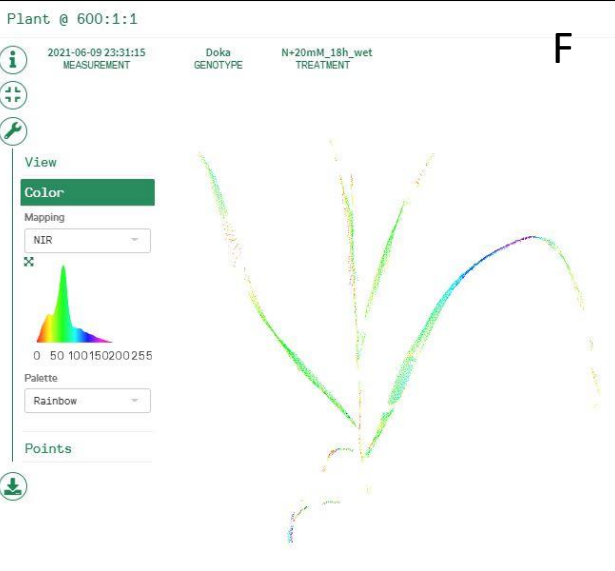


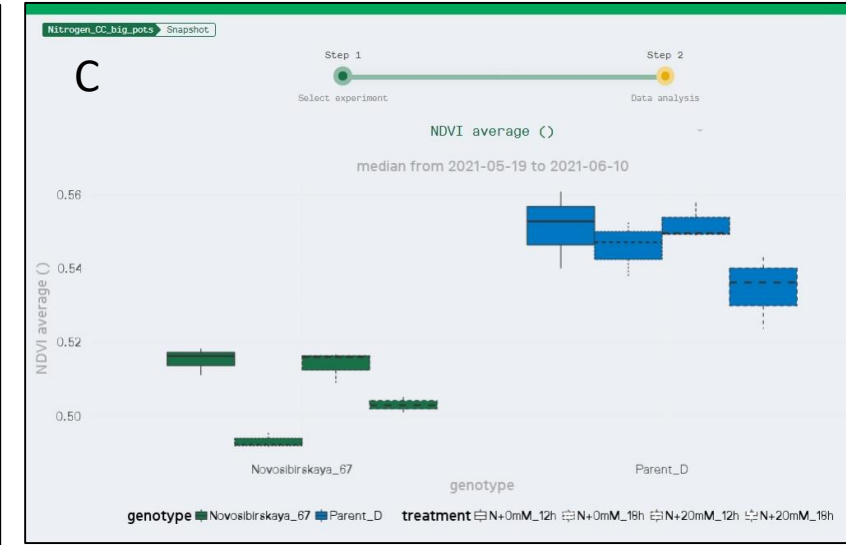
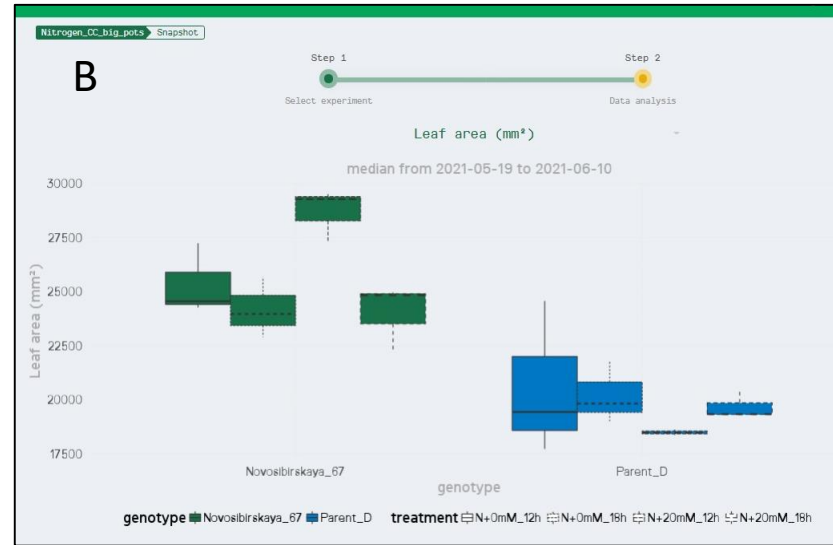
**Figure 5.** Kompetitive allele specific PCR (KASP) genotyping for *Rht1*.

Specific experimental culturing conditions causes stress and changes in wheat leaves that are detected using spectral capability of multispectral digital phenotyping system TraitFinder (Phenospex, Netherlands)



**Figure 6.** An *Rht17(Rth-B1p)* wheat plant demonstrate signs of stress as can be seen by eye (A) and how it is recognized by the phenotyping system (B,C). Figures D-G show reflections of the plant A in 4 spectral channels: Red, Blue, Near infrared and Green respectively. These spectral data are transformed to spectral indices – NDVI (as shown in panel H for different wheat lines).





**Figure 7.** The response of two different common wheat lines to various growing condition. A: dynamics of growth of the leaf area; B: leaves of the first wheat line (on the left) grow faster and stronger respond to the growing conditions; C: the second wheat line exceed the first one in photosynthetic capability that is reflected in the NDVI plot.

## Conclusions

New markers for *Grf* genotyping have been developed. This opens up new horizons in MAS for breeding new plants with the most effective combinations of *Grf* and *Rht* alleles. The digital phenotyping is useful tool for revealing growing and spectral patterns during the whole lifespan of the plant. The associations between these patterns and valuable traits holds new promise for the breeders.