Changing the odds in our favor: Manipulating the microflora in wheat to reduce *Stagonospora nodorum* blotch

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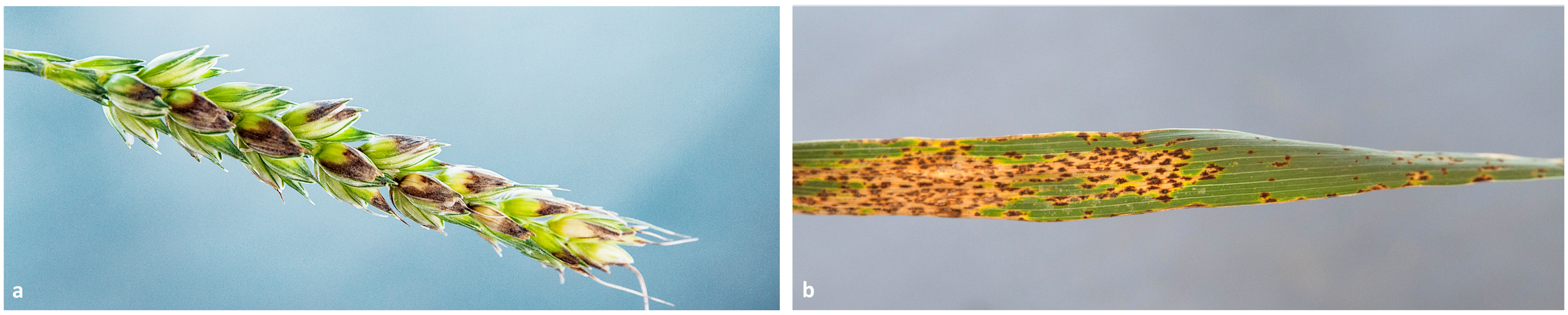

Figure 1. *Stagnospora nodorum* blotch (SNB) on a) heads and b) leaves of wheat. Photo: Andrea Ficke

INTRODUCTION

Recently, the Norwegian Institute for Bioeconomy (NIBIO) has launched a five year strategic institute project in order to encourage inter-disciplinary work between plant pathology, genetics and bioinformatics.

Our work group is in particular interested in understanding what factors are tipping the balance of the microbial community in favor of the common wheat diseases, such as *Stagonospora nodorum* blotch (SNB, Fig. 1 a/b), and how we could manipulate this balance. Several investigators found that the microbial community was significantly influenced by plant organs, sampling sites and growth stage of the plant (Siebert *et al.* 1988, Crous *et al.* 1995, Larran *et al.* 2007, Vujanovic *et al.* 2012, Grudzinska-Sterno *et al.* 2016). These studies suggested that agronomic practices can strongly influence the phytobiome of wheat. However, how the phytobiome is changing under biotic stresses, such as plant diseases, has not been investigated.

We wish to determine the microbial community in wheat as it changes with a) SNB infection, b) Cultivar, c) Growth stage, d) Plant organ and e) Application of plant protection products and how these factors interact.

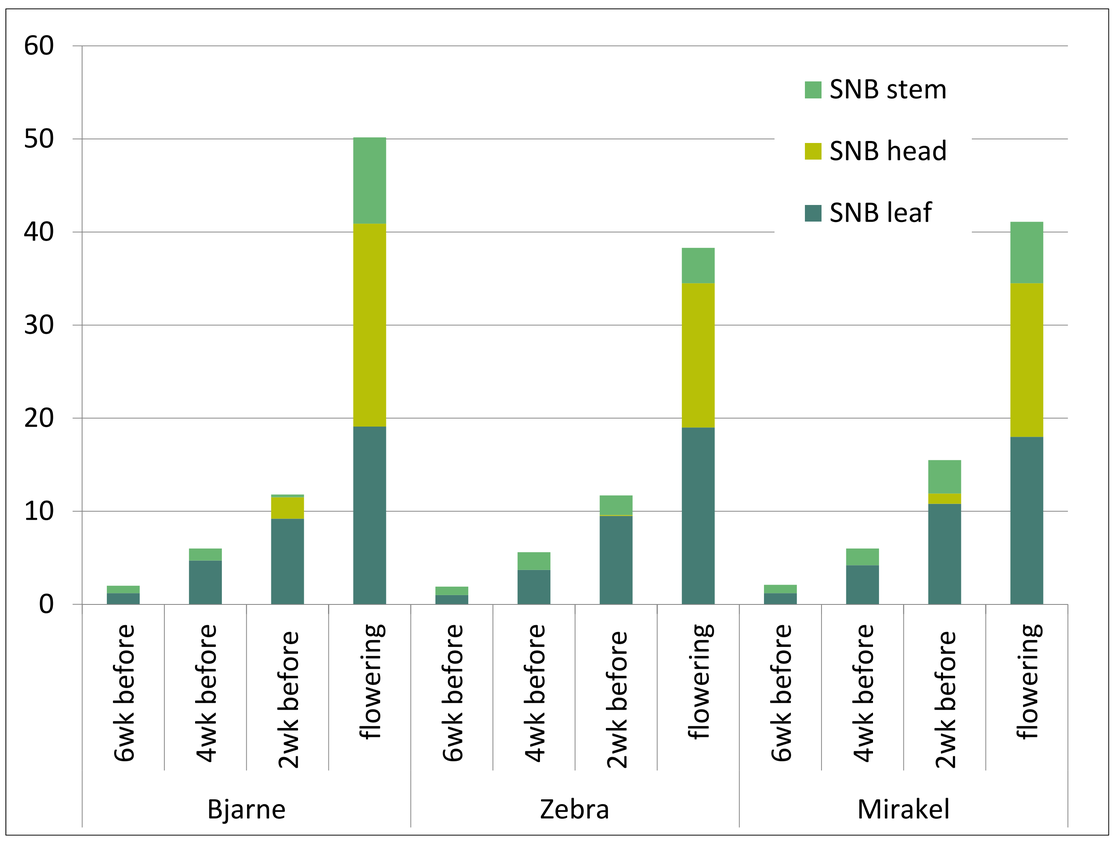

Figure 2. Disease severity (%) of SNB on stems, heads and leaves of 'Bjarne', 'Zebra' and 'Mirakel' inoculated at flowering and two, four, six weeks before flowering. Assessed two to three weeks after inoculation.

MATERIALS AND METHODS

The first greenhouse studies were conducted in 2017 by sowing three different spring wheat cultivars ('Bjarne', 'Zebra' and 'Mirakel') in two-week intervals (five plants per pot, 24 pots per cultivar per sowing, four sowing times) under a 16h/8h light/dark, 55% RH and 15C/18C night/day regime. Pots were completely randomized after every new sowing.

We inoculated 12 pots per cultivar per growth stage with 10^6 spores/ml *P. nodorum* suspension (1% Tween 20) when the oldest plants were flowering (GS 60-65) and repeated the inoculation after two weeks.

Control plants were sprayed with 1% Tween 20 in water only. All pots were bagged with clear plastic bags to increase humidity to 100% for 48hr. Plants were assessed for disease severity per pot, leaf, stem and head two to three weeks after inoculation.

We harvested two plants and divided them in stems, leaves and heads and kept them at -20°C for DNA extraction and sequencing.

Ten field trials in the South-East of Norway were used in 2017 to collect disease data, nutritional status of the plant and leaf samples from 'Bjarne', 'Zebra' and 'Mirakel' at GS 60-65 and GS 70-75.

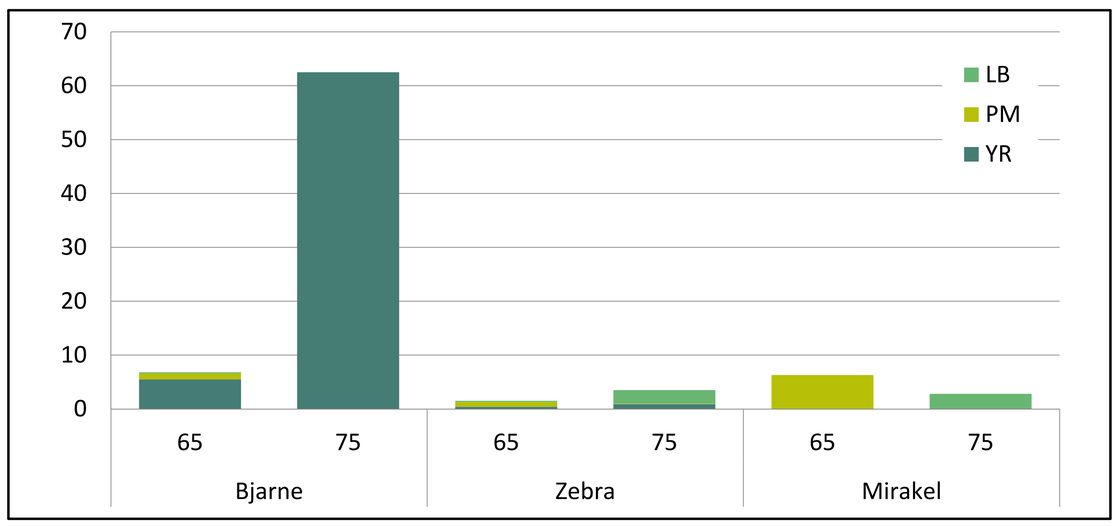

Figure 3. Disease severity (%) of leaf blotch (LB, including SNB), powdery mildew (PM), and yellow rust (YR) on spring wheat cultivars 'Bjarne', 'Zebra' and 'Mirakel' assessed in the field at GS 65 and 75.

Leaf samples will be used for sequencing to determine the microbial community present at the different growth stages and linked to the observed severity of different wheat diseases.

PRELIMINARY RESULTS

In the greenhouse study, we observed clear differences in SNB development. SNB was positively correlated with the growth stage at inoculation times (Fig. 2). Even though, all plant organs tested were susceptible, the increase in susceptibility was strongest in the stem. There were no significant differences between cultivars. However, the field, the different cultivars displayed a clear difference in susceptibility to the different diseases, especially yellow rust (Fig. 3).

FUTURE WORK

This project is intended to increase our expertise in the design of sequencing pipelines, analysis of complex molecular data and interpretation in the field of multitrophic interactions from the molecular to the farm level. We are in the process of isolating DNA from the tissue samples from the greenhouse experiment and the field trials. We are currently developing a sequencing pipeline for bacterial, fungal and oomycete 16S, ITS and other loci to determine the identity and quantity of the different microbial organisms. We will sample more field trials of the same three cultivars to investigate the effect of fungicide treatments, location and weather on the microbial community and the plant health status in the next three years.

References

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