



ANTIFUNGAL-PRODUCING BACILLACEAE STRAINS ENHANCE WHEAT YIELD AND BAKING QUALITY IN FIELD CONDITIONS

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Regenerative agriculture has emerged as a sustainable alternative, promoting environmentally friendly practices aimed at maintaining or restoring soil health over the medium and long term. A key aspect of this approach involves the use of microbial-based bioinputs to reduce or replace agrochemical inputs, targeting both nutrient management and pathogen control. This influences the global biological market, which was valued at \$14.6 billion in 2023 and is projected to reach \$27.9 billion by 2028 (Frederick, A and Mei 2024).

Figure 1. Evaluation of bacterial supernatant antifungal activity in wheat seeds.

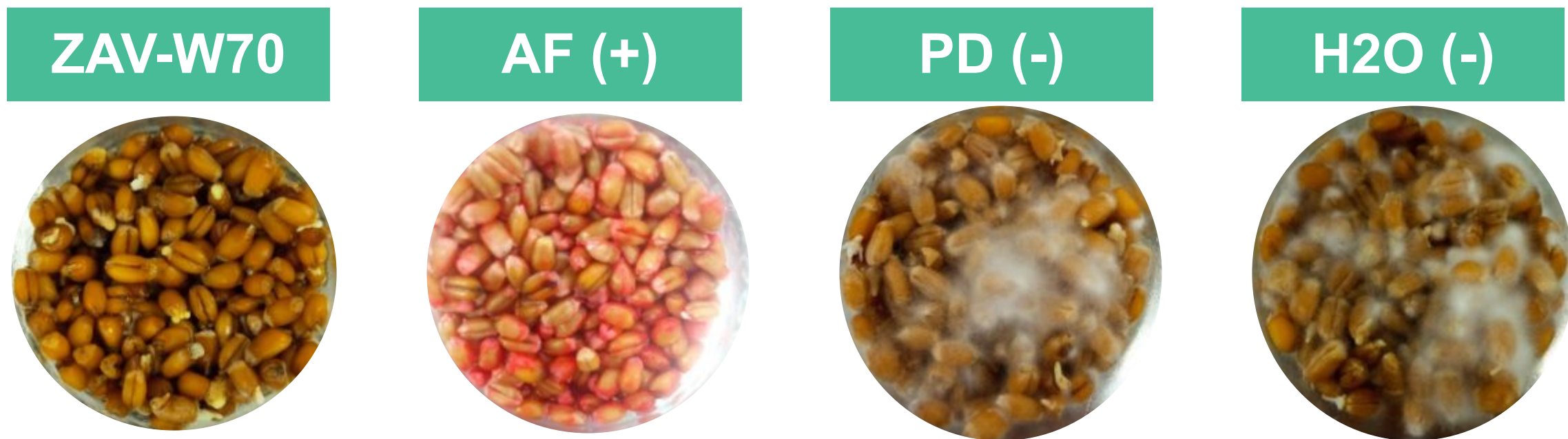


Table 1. Activities of the selected isolates associated to biocontrol and/or plant growth promotion detected by in vitro assays.

	ZAV-W70	ZAV-W64
IAA*	0.62 ± 0.47	0.04 ± 0.04
Phosphate solubilization	++	+
Siderophores	+++	+
Biofilm formation*	1.89 ± 1.75	0.243± 0.10

Qualitative results are indicated as + for positive (+++ high, ++ medium o + low intensity) or (-) for negative results. *Absorbance values.

ZAV-W70 acts as both a growth promoter and a fungicide: ZAV-W70 treatments led to increased plant height and a higher number of fertile spikes in treatments without fungicide. ZAV-W64 with fungicide also increased yield by 7% compared to the water control. This yield improvement was associated with a slight increase in the number of fertile spikes, while grain weight remained similar to the control.

Seed treatment with ZAV-W64 improved bread-making quality, evidenced by significant increases in total gluten content (10%) and alveograph W parameter (15%) compared to fungicide treatment alone (Fig. 4).

The increase in total gluten for ZAV-W64 with fungicide correlated with higher protein content. Moisture levels were similar across treatments, while water and fungicide treatments showed increases in ash content and test weight, respectively.

CONCLUSIONS

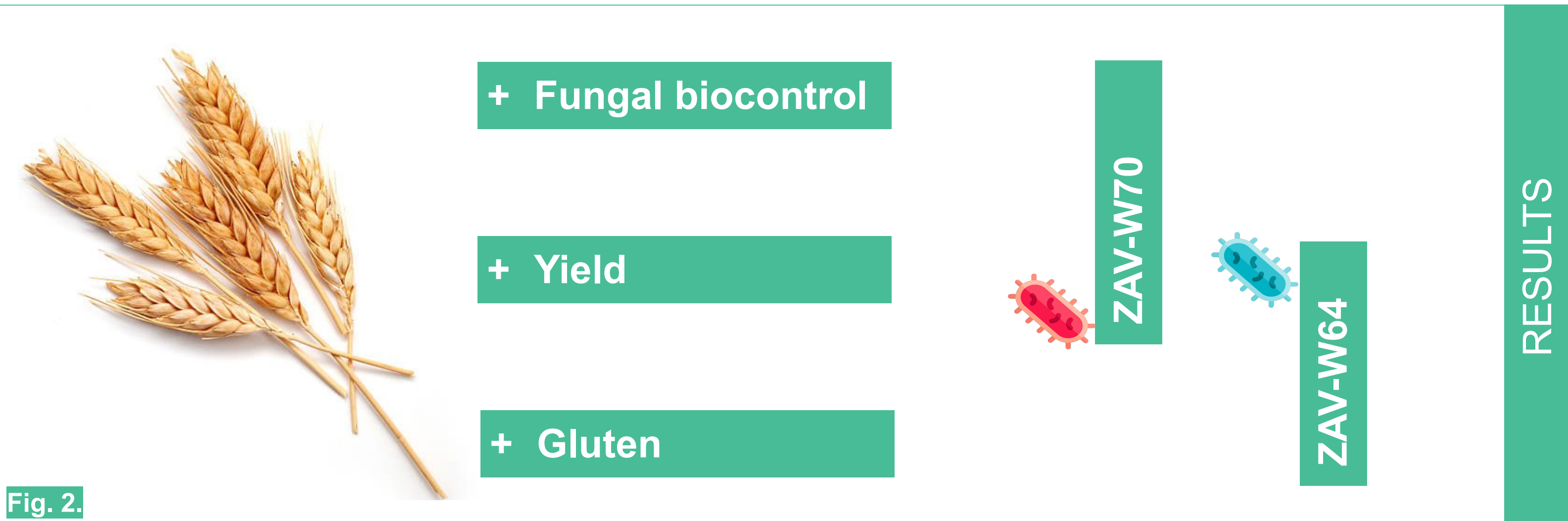
The degradation of soil microbial diversity driven by agricultural intensification underscores the urgent need for sustainable alternatives. These findings support the potential of beneficial rhizobacteria to contribute to more sustainable wheat production systems.

Table 1. Materials and methods. Field trials.

Seed treatments	T1 (water control), T2 (ZAV-W64), T3 (ZAV-W70), T4 (fungicide, Metalaxyl®), T5 (fungicide + ZAV-W64), and T6 (fungicide + ZAV-W70).
Strain growth conditions	The ZAV-W64 and ZAV-W70 strains were grown in potato-dextrose medium and formulated to a concentration of $\geq 10^8$ spores mL ⁻¹ (Navira SA).
Year and location of field trial	2022: Unidad Experimental de Cultivos Extensivos of the Facultad de Ciencias Agrarias (UNL) in Esperanza, Santa Fe. Typical Argiudol soil from the Esperanza series. Field plots, measuring 3 meters wide by 10 meters long, were sown using an experimental seeder with a row spacing of 17.5 cm. The short-cycle wheat variety Nutria (Klein seedbed) was sown on July 12, 2022. 2023: Estación Experimental del INTA in Oliveros, Santa Fe, on a typical Argiudol soil from the Maciel series. Four field plots per treatment were arranged in a complete block design, each measuring 3 meters wide by 12 meters long. The intermediate-cycle wheat variety Buck SY-109 (Buck seedbed) was sown on June 22, 2023, using an experimental seeder with a row spacing of 20 cm.

For bacterial treatments, 20 mL of inoculant per kg of seeds was applied by manual shaking in a plastic bag. For fungicide treatments, 2 mL of Metalaxyl® solution (10 µg mL⁻¹) per kg of seeds was used. The crop was maintained free of pests (insects and weeds) and diseases. At physiological maturity, plants from the central rows covering an area of 1 m² were manually harvested and threshed using an experimental thresher. Statistical analyses were performed using InfoStat software, version 2020. The effects of treatments on kernel antifungal capacity were evaluated using a unifactorial ANOVA (treatment factor), with mean comparisons conducted using the LSD test. Field assay data were analyzed using a two-factor mixed model ANOVA (treatment and fungicide factors), with mean comparisons made using the LSD test. The assumptions of ANOVA, including normal distribution of residuals (assessed using the Shapiro-Wilk test and QQ plots) and homoscedasticity (evaluated using the Levene test and residual plots), were verified for all analyses. For variables that did not meet ANOVA assumptions, non-parametric Kruskal-Wallis tests were performed, with data partitioned by assay.

To explore alternatives to chemical fungicides, we evaluated the effects of *B. velezensis* ZAV-W70 and *P. megaterium* ZAV-W64 in wheat field trials.



ZAV-W70 alone achieved yields comparable to those obtained with chemical fungicide co-application and increased yield by 11.7% relative to the water-treated control (Fig. 3).

Figure 3. Effects of ZAV-W64 and ZAV-W70 on wheat yield under field conditions. Yield values are expressed as means ± SEM (n = 3 for 2022; n = 4 for 2023) and measured at physiological maturity. Bars not sharing the same letter are significantly different according to ANOVA and LSD multiple comparison test (p < 0.05). The overall average yield was 2446.1 kg ha⁻¹, with maximum yields of 2584.3 kg ha⁻¹ (fungicide and ZAV-W70 combination) and minimum yields of 2298.3 kg ha⁻¹ (water control).

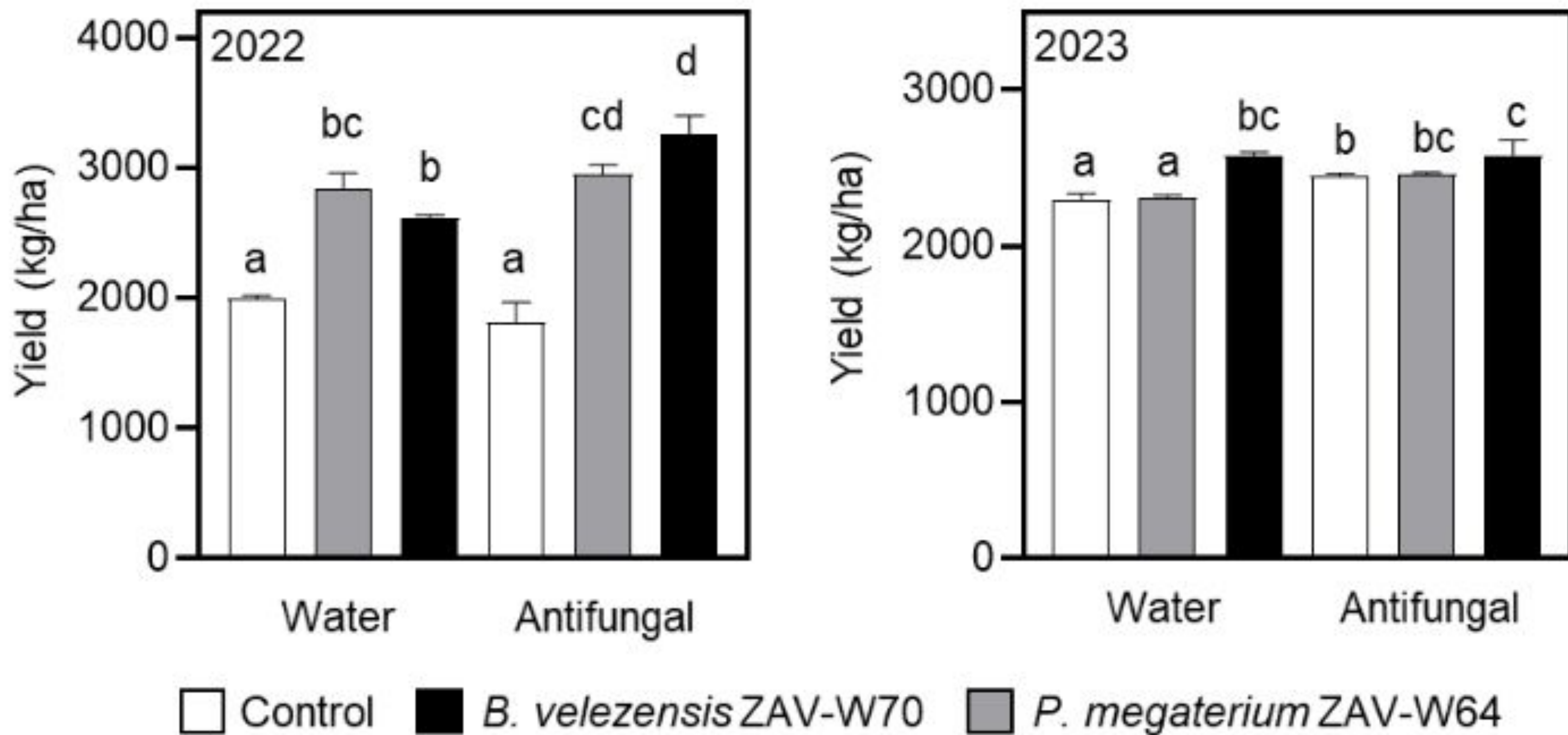


Figure 4. Effects of ZAV-W64 and ZAV-W70 on wheat seed quality parameters. Wet gluten content (left panel) and alveograph W parameter (right panel), indicating dough strength, in seeds produced from plants grown from seeds treated with ZAV-W64, ZAV-W70, or controls (water and fungicide) prior to sowing. Values are expressed as means ± SEM (n = 3). Bars not sharing the same letter are significantly different according to ANOVA and LSD multiple comparison test (p < 0.05).

