GBI-WP2- OP 2.6: Validation of heat treatment in combination with chemicals protocol for bacterial elimination in rice seed

Category: Innovation Development

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Summary: A major concern in the exchange of rice germplasm is the potential contamination of seeds with bacterial species. This contamination could have an effect not just on rice crops but also on other crops that are economically important. The integration of heat treatment with the use of fungicide is a viable and economical option that potentially targets both bacterial and fungal pathogens. In 2024, we carried on with heat treatment experiments, exposing seeds to temperatures between 50 and 70˚C over different durations ranging from 3 to 20 days. Our study demonstrated that heat treatment at 60°C for three days effectively eliminated between 80 and 90% of the bacteria, offering a viable alternative to the use of bactericides, particularly when paired with fungicide (Teams) treatment.

Challenge: Unintentional exchange of rice seeds contaminated with bacteria is a common occurrence since the bacteria hide in the embryo. Moreover, the limited availability of bactericides in many laboratories, coupled with concerns regarding safety, antibiotic resistance, and cost-effectiveness in their use, poses a considerable challenge for most labs. Consequently, the use of bactericides for seed treatment may not be sustainable. Thermotherapy is an economical approach, as most bacteria are eliminated through heat treatment when subjected to a sufficiently extended duration. However, its necessary to optimize the temperature at which bacteria are eliminated while maintaining seed viability.

Methodology:

Infected rice seeds from four rice varieties were exposed to three different temperatures (50, 60, and 70°C) to assess the impact of heat treatment on elimination of bacteria from contaminated seeds. Additionally, temperature treatment was combined with four chemicals to assess if the combination of heat treatment and fungicide treatment would effectively eradicate bacteria. The seeds underwent heat treatment both with and without the application of seed dressing chemicals over a duration of 3, 5, 10, and 20 days. Following a specified duration, the seeds were plated in sterile PSA media to assess the growth of bacteria. Data on the survival rate of bacteria were submitted to linear mixed effect models to assess the variation of bacterial survival rate according to the experimental factors (treatment with 4 levels, variety with 4 levels, temperature with 3 levels and day with 5 levels). In this model, all factors were considered as fixed whereas replication from design of experiment was considered as random. Model fitting was done using asreml-4 (The VSNi Team, 2023) in R statistical environment (R Core Team, 2024).

Results

The results of the experiments involving a combined heat and chemical treatment are presented in Figure 1 & 2. All varieties of infected rice seeds showed significant sensitivity to heat treatments, leading to a decrease in germination rates, particularly at 70°C. The application of heat treatment at 60°C for a duration of three days demonstrated significant efficacy, effectively eliminating bacteria by 80 to 90%. The heat treatment at 70°C demonstrated greater efficacy, but it had a significant impact on seed germination. Overall, heat treatment presents a promising alternative to bactericides, especially when combined with fungicide treatment (Teams).

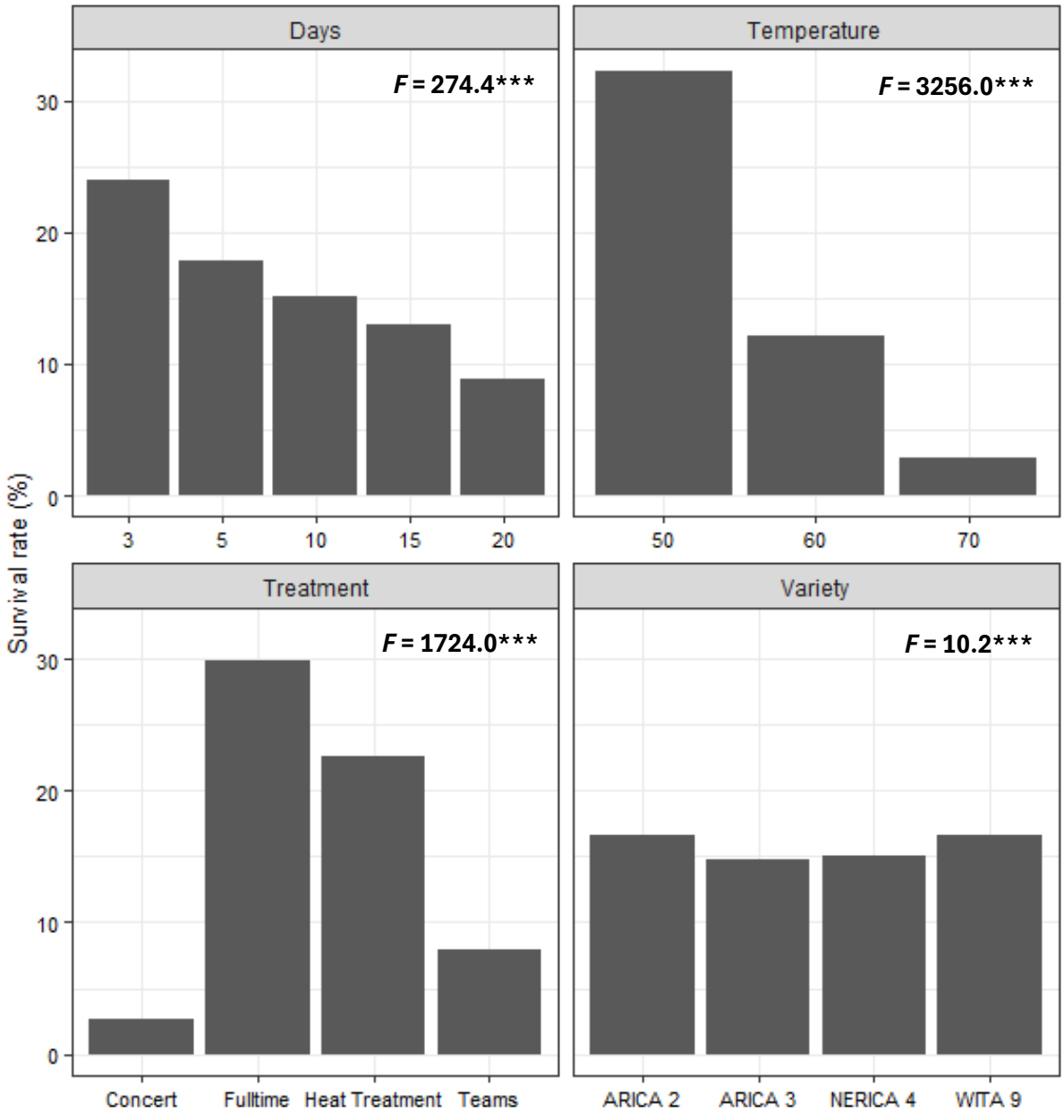

Figure 1. Main effect of experimental factors on survival rate

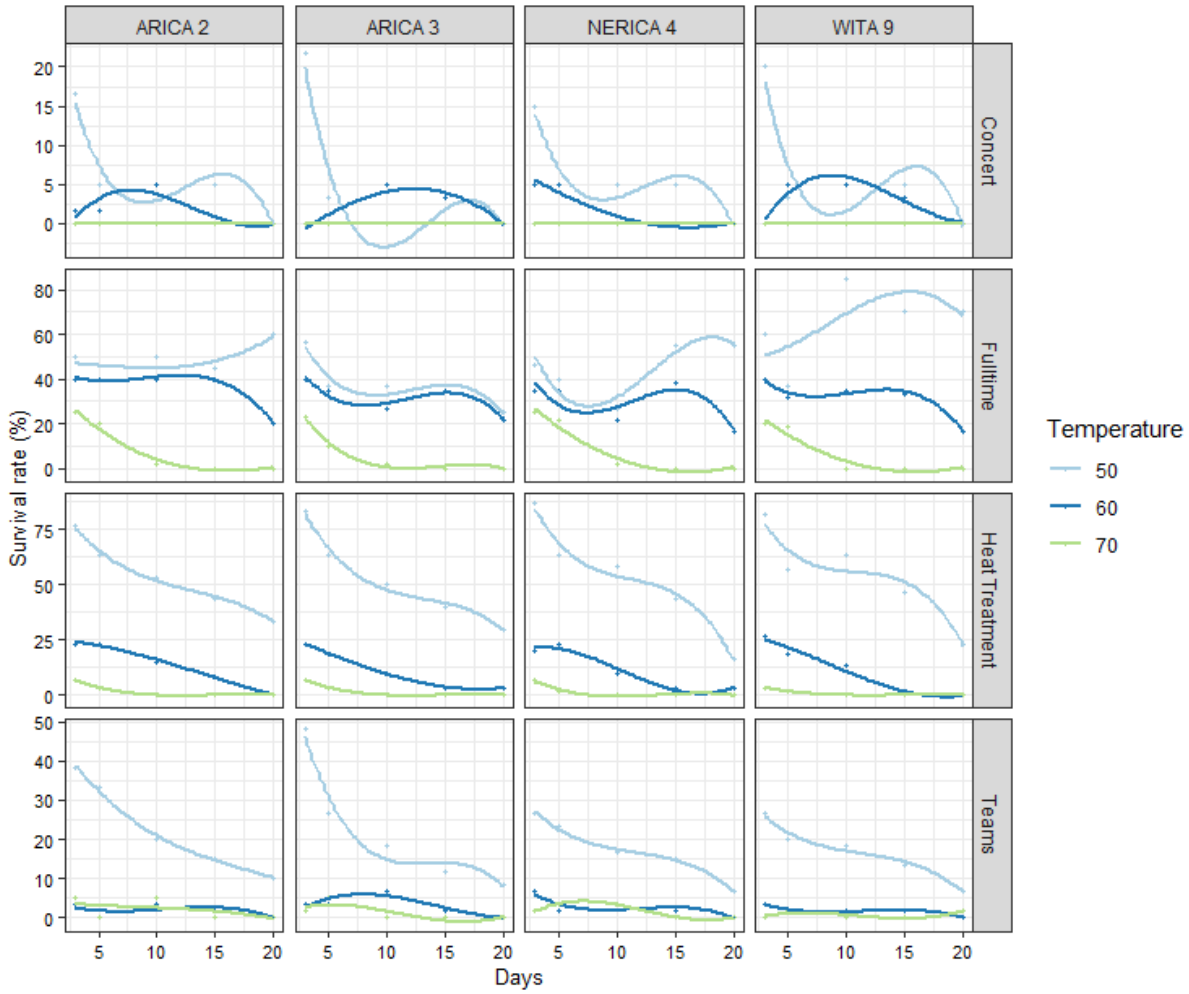

Figure 2. Variation of survival rate according to the third level significant interaction

References

R Core Team (2024). R: A Language and Environment for Statistical Computing_. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org.

The VSNi Team (2023). asreml: Fits Linear Mixed Models using REML. R package version 4.2.0.355, www.vsni.co.uk.