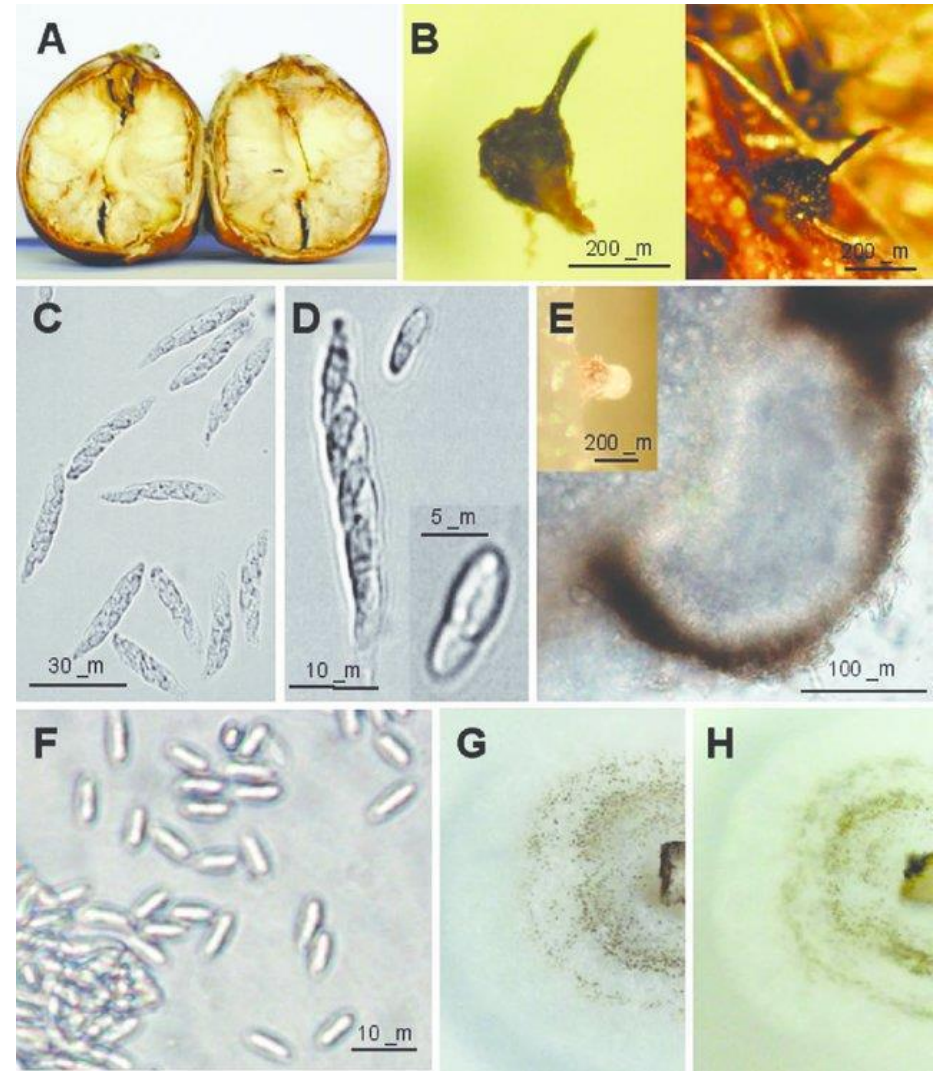


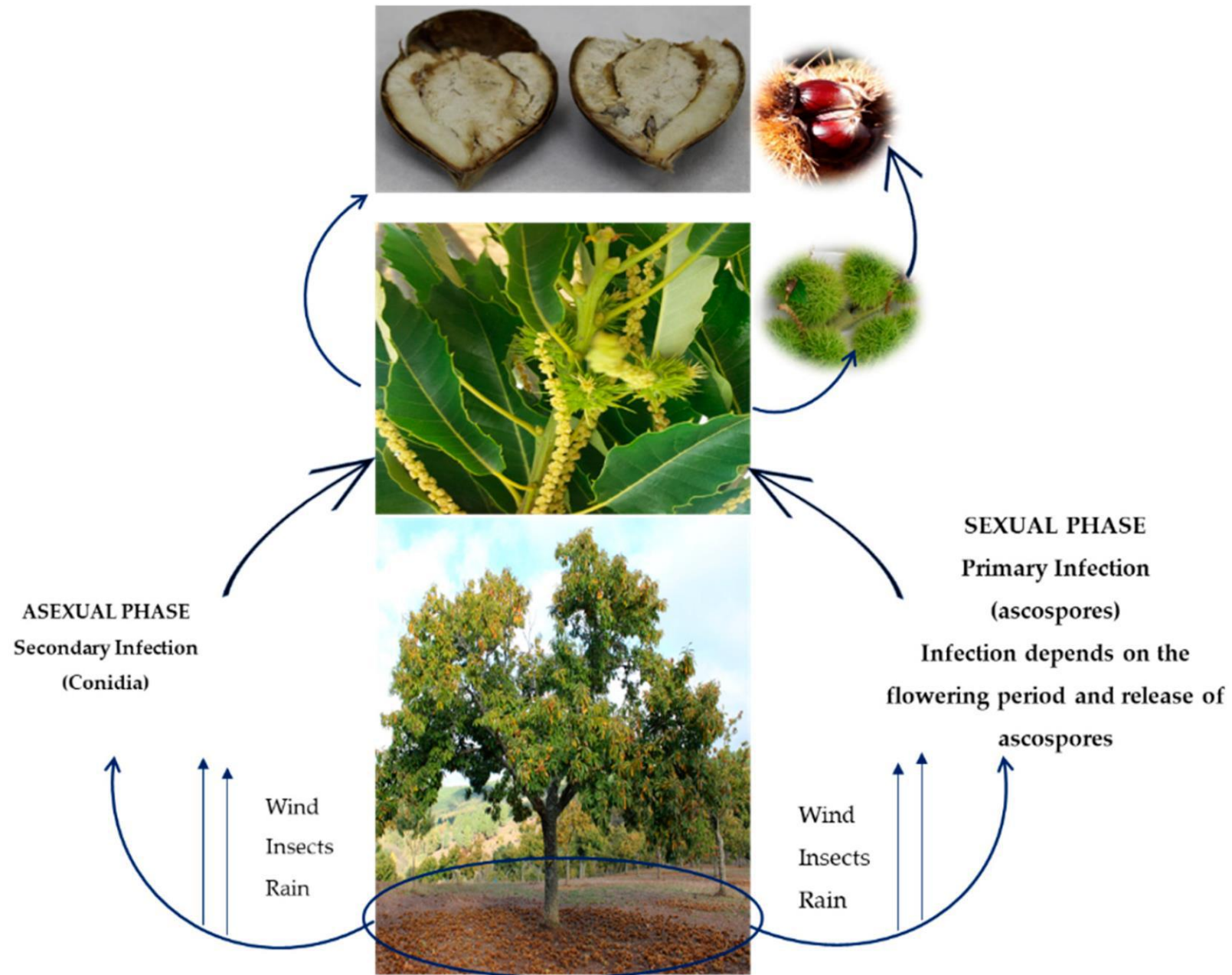
Nut Rot



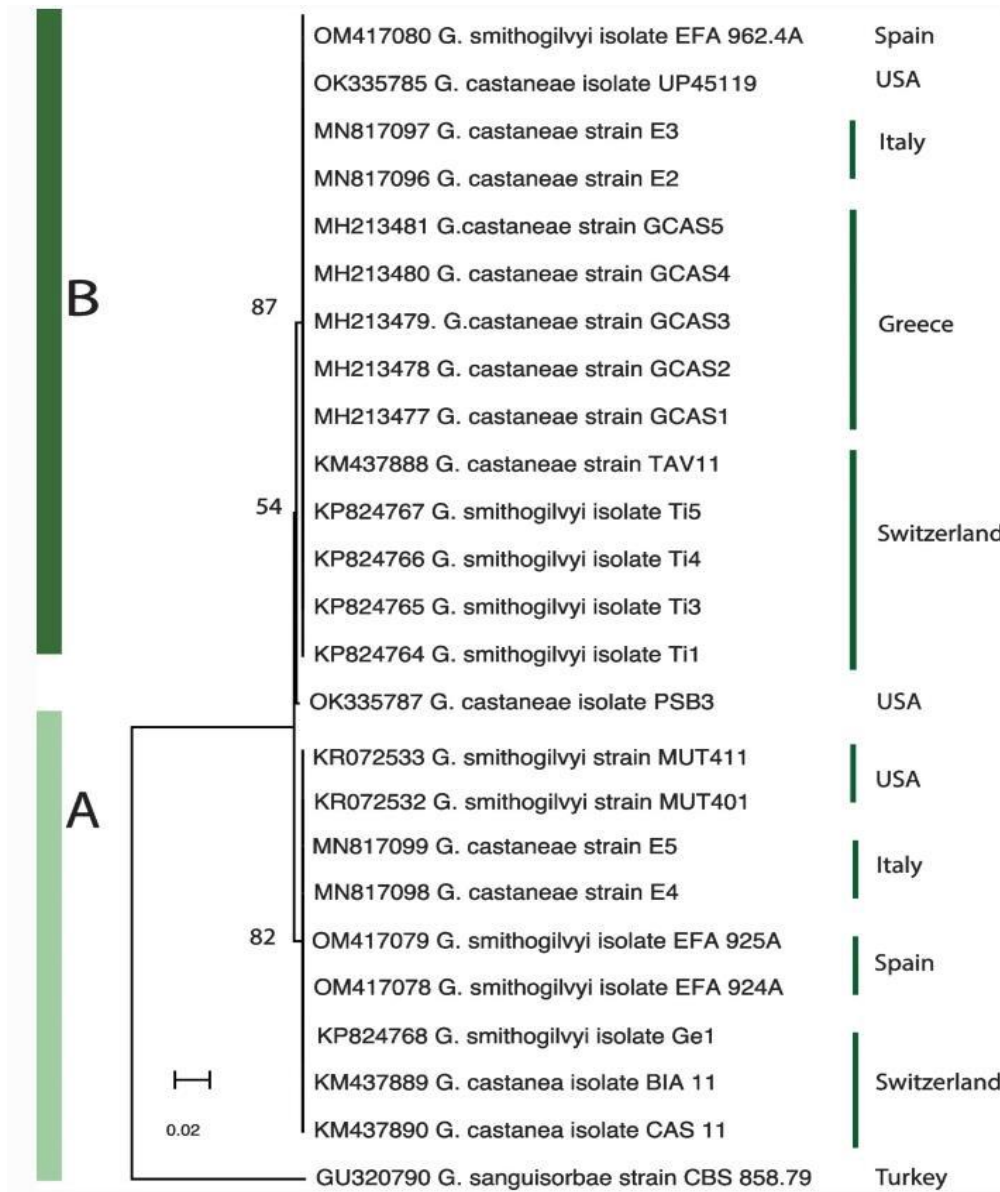
Celeste Linde

Gnomoniopsis smithogilvyi
Syn *G. castanea*





Infected chestnuts and burrs (primary inoculum). Infected leaves (fall in autumn/winter). Infected branches remain latent in winter.



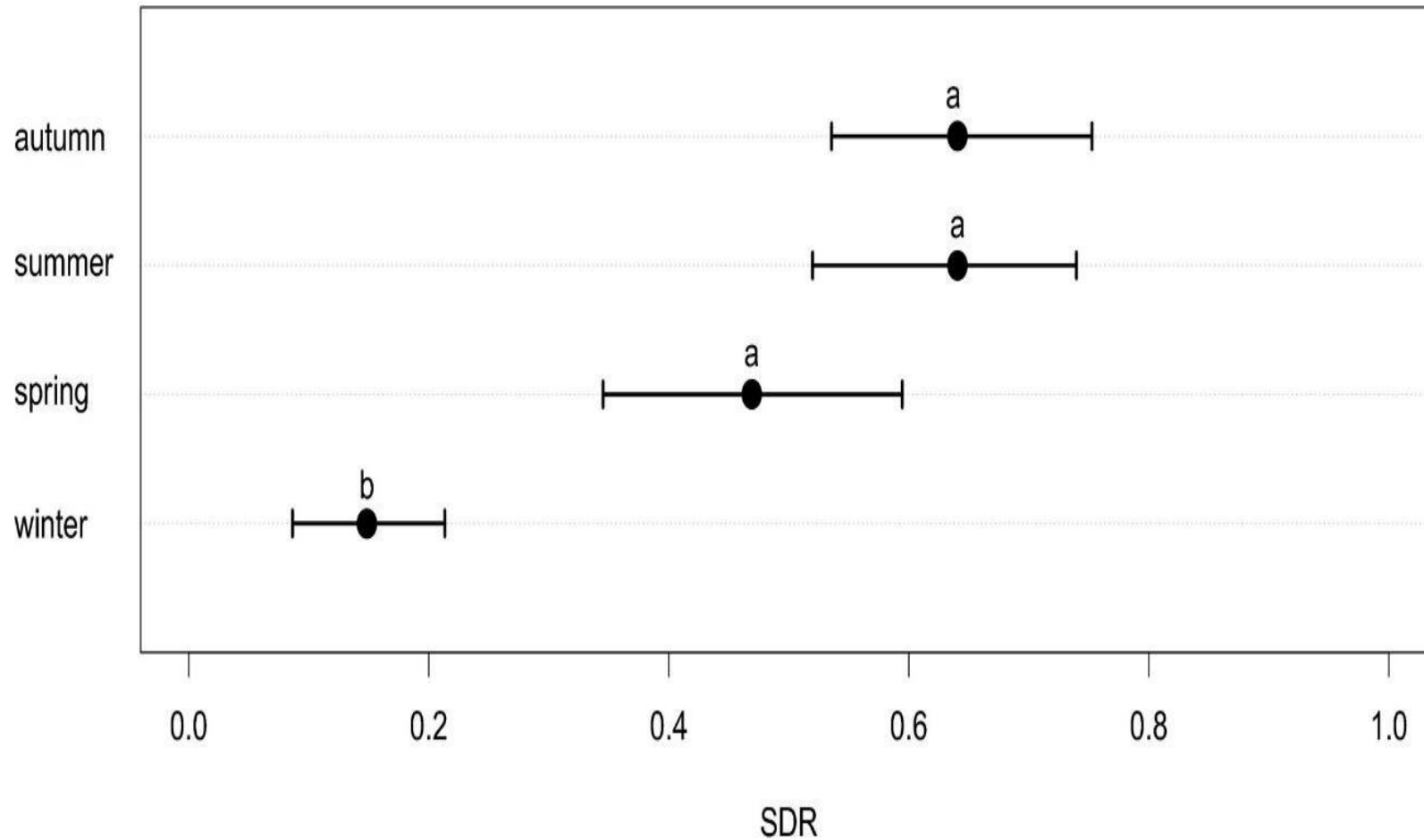
Gnomoniopsis smithogilvyi β -tubulin neighbour-joining tree. This figure is reproduced from Seddai et al. (2023). Two haplotypes (A and B) within *G. smithogilvyi* are indicated.

Factors associated with disease

Although rainfall is critical for ascospore formation, temperature is reported to play the more important role in disease development.

Based on modelling various climatic factors, **an increase in temperature in the months before nut harvesting** was reported to lead to a higher incidence of nut rot in northwest Italy.

Based on a spore trapping experiment over 2 years, spore (both ascospores and conidia) deposition occur all year round, but mostly in summer and autumn (Lione et al. 2021)



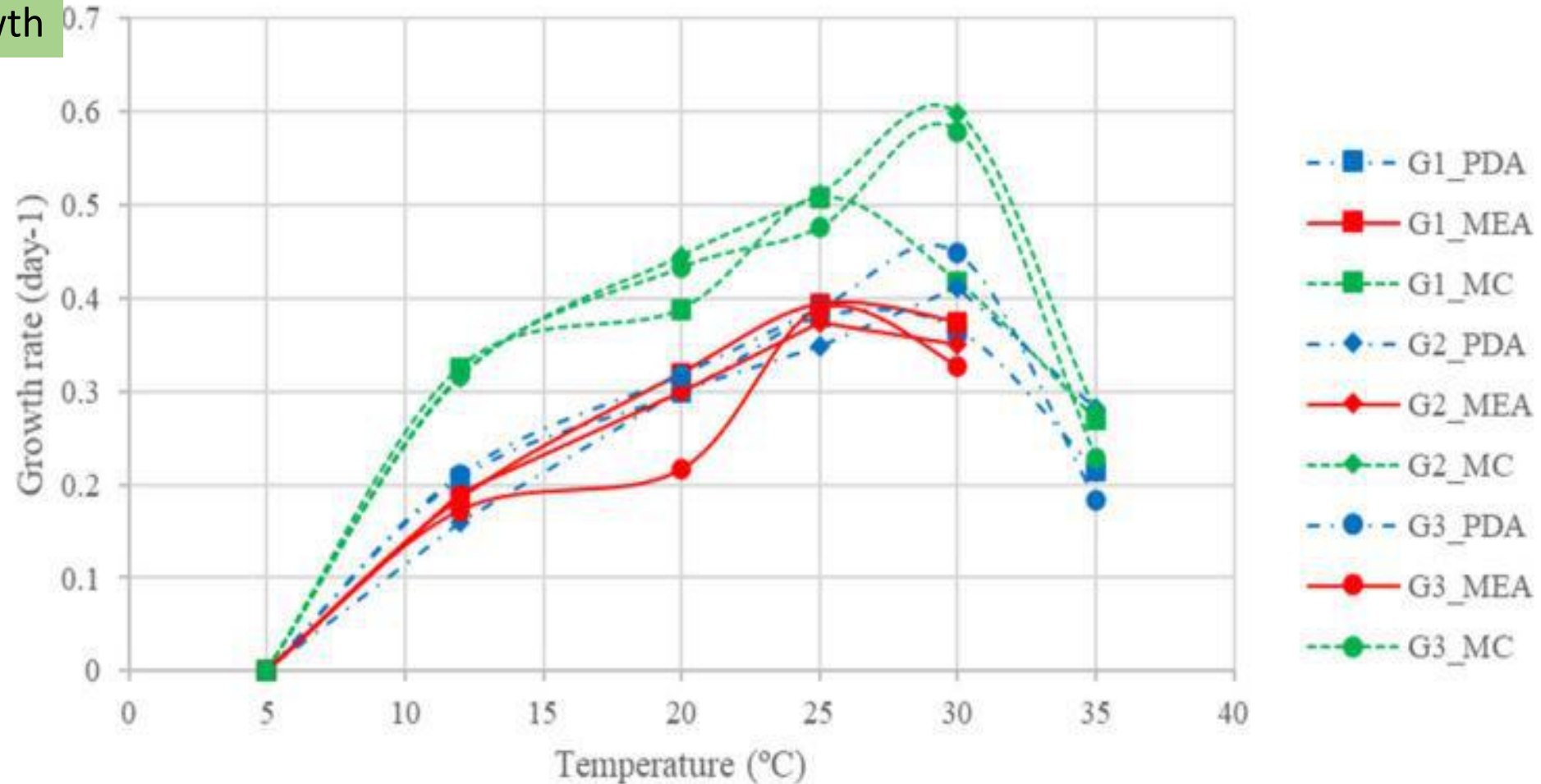
High temperatures – climate change

Spore deposition was positively correlated with mean, maximum, and minimum temperature, growing degree days at 0 and 5 °C thresholds, and wind gusts, for the summer to autumn months. This suggests that **warmer temperatures in the second half of the vegetative season** are associated with increased nut rot incidence.

It is expected that **rising temperatures** and **strong winds** due to **climate change** may increase the incidence of the disease in the future.

Although there is no correlation between disease incidence and rainfall in the modelling study, **drought** may still play a role in predisposing trees to infection

Culture growth



Growth of *Gnomoniopsis smithogilvyi* isolates G1 – G3 in potato dextrose agar (PDA), malt extract agar (MEA) and chestnut medium (MC) at different temperatures. Figure reproduced from Possamai et al (2023). The green lines show all three *G. smithogilvyi* isolates growing best on chestnut medium. On this medium isolates grow best at 30 -35 °C.

Conidial production on different media at different temperatures

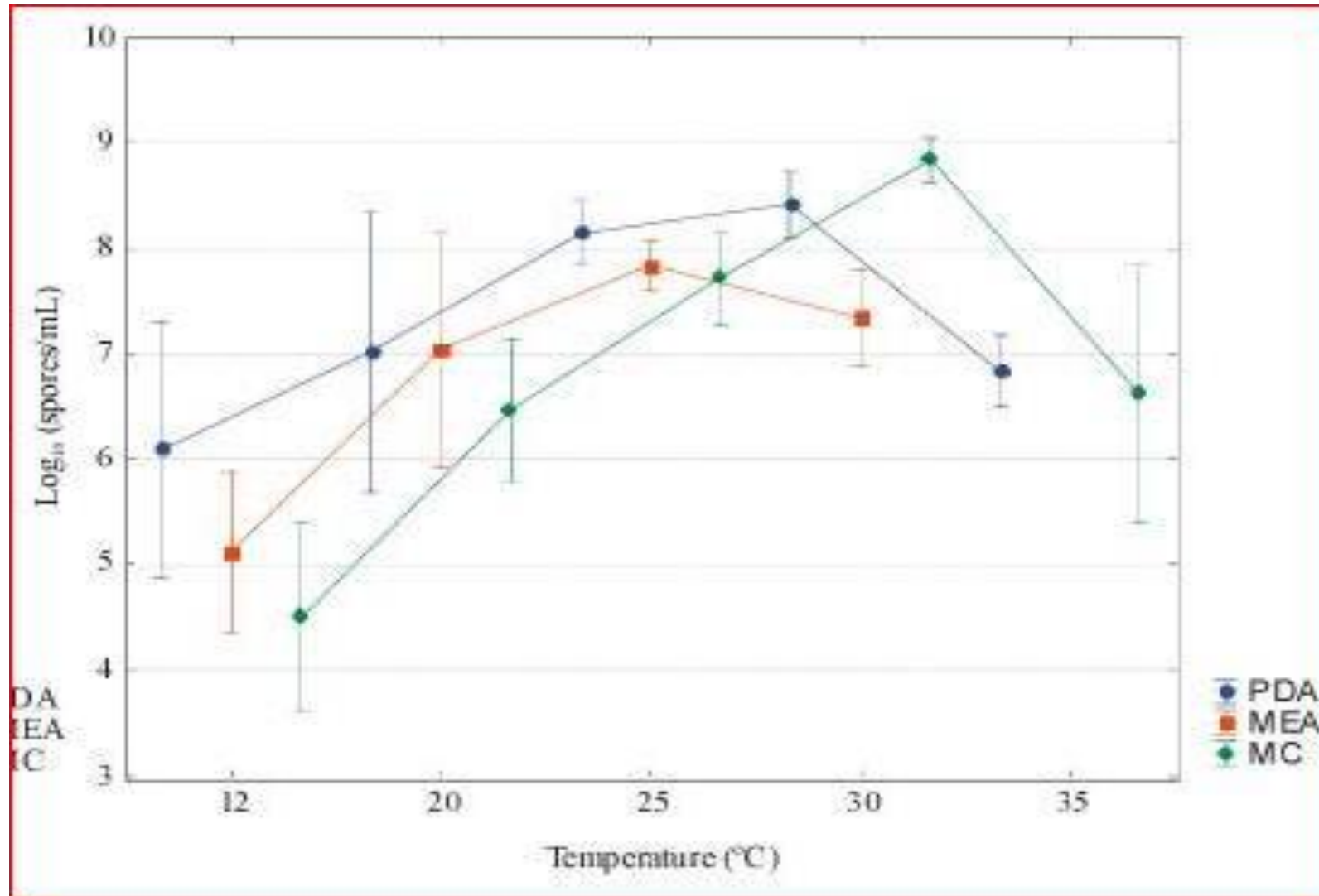
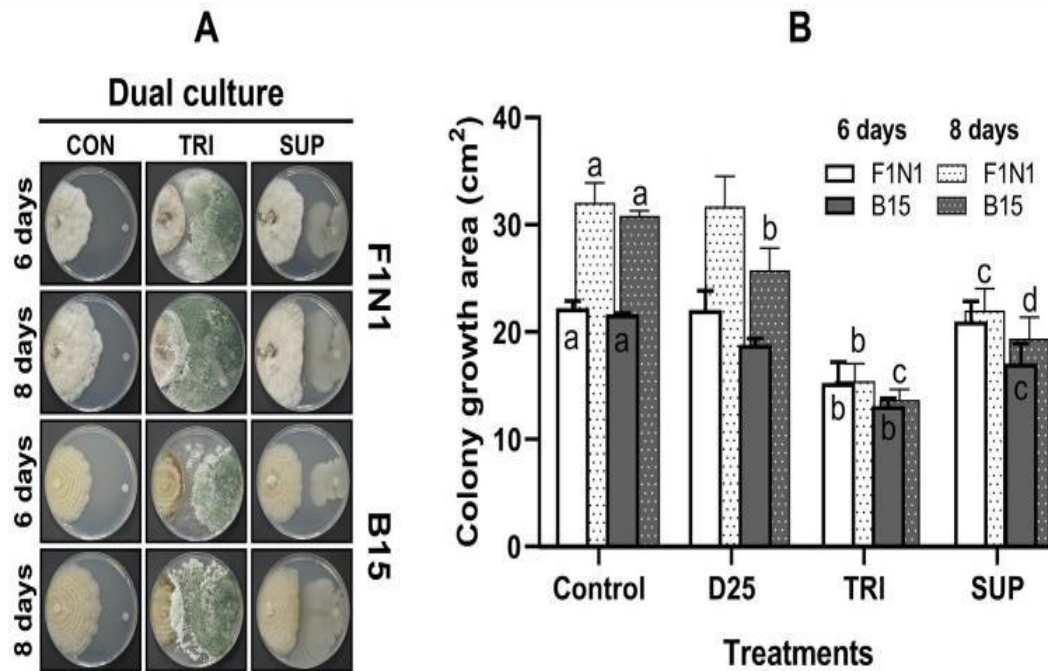


Figure reproduced from Possamai et al (2023). This graph shows that not only is conidial production highest on chestnut medium (MC), but also that most conidia are produced at 30 °C.

Control of nut rot: Biological control

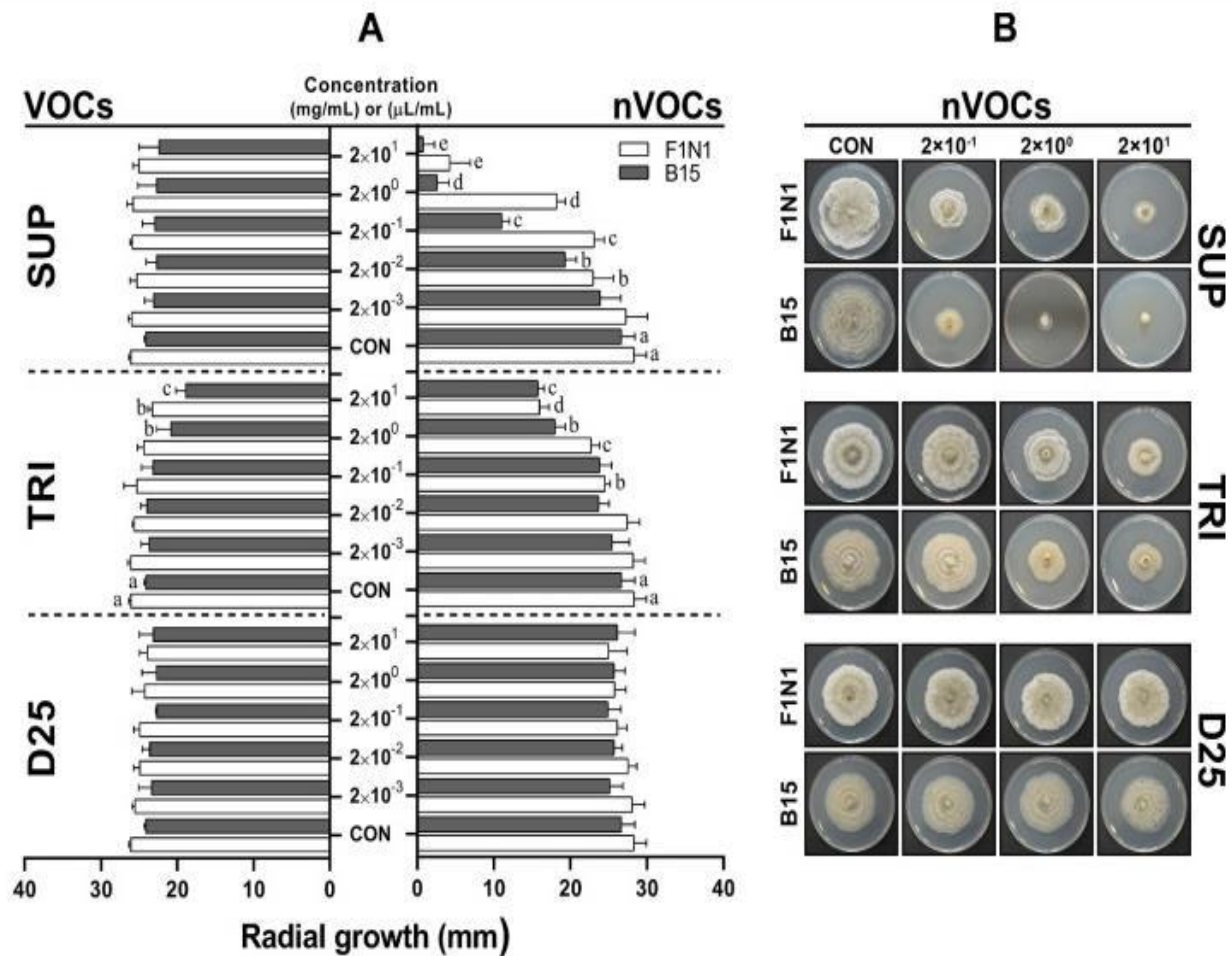


TRI = Trichoderma
 SUP = *Bacillus*
 D25 = ?

Trichoderma (TRI) is most effective at suppressing growth of *G. smithogilvyi*

Figure reproduced from Silva-Campos et al (2022).

Effect of the biological control agents TRI and SUP on the growth of *G. smithogilvyi* isolates B15 and F1N1 evaluated in a dual culture assay. **A** Inhibition of growth of *G. smithogilvyi* due to exposure to TRI and SUP at 6 and 8 days in the dual culture assay compared with controls. Note for the BCA present in TRI overgrowing *G. smithogilvyi* colony and the halo of inhibition displayed under the SUP treatment. **B** Growth area of the isolates measured after 6 and 8 days. Means \pm SEM labelled with different letters are significantly different to the control according to Dunnett's test at $p = 0.05$

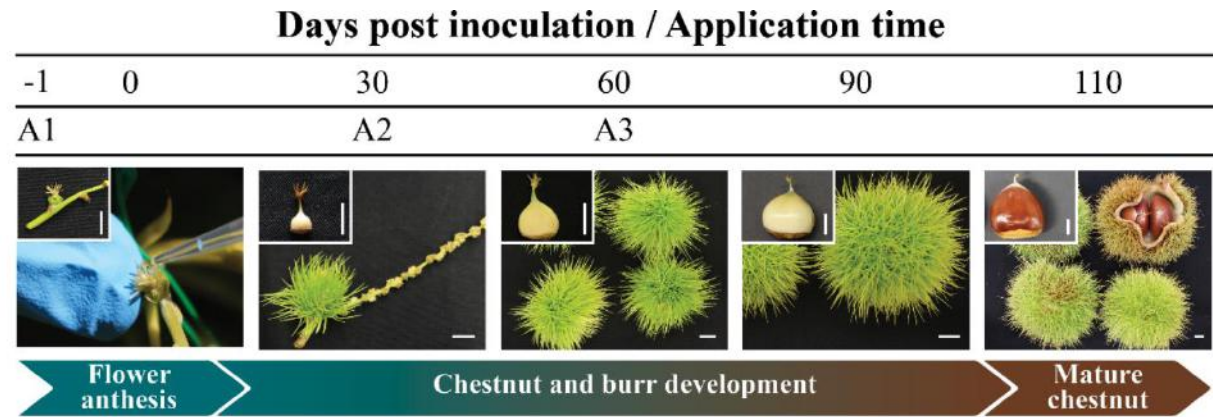


Non-volatiles (metabolites) from TRI and SUP provide some suppression of *G. smithogilvyi* growth.

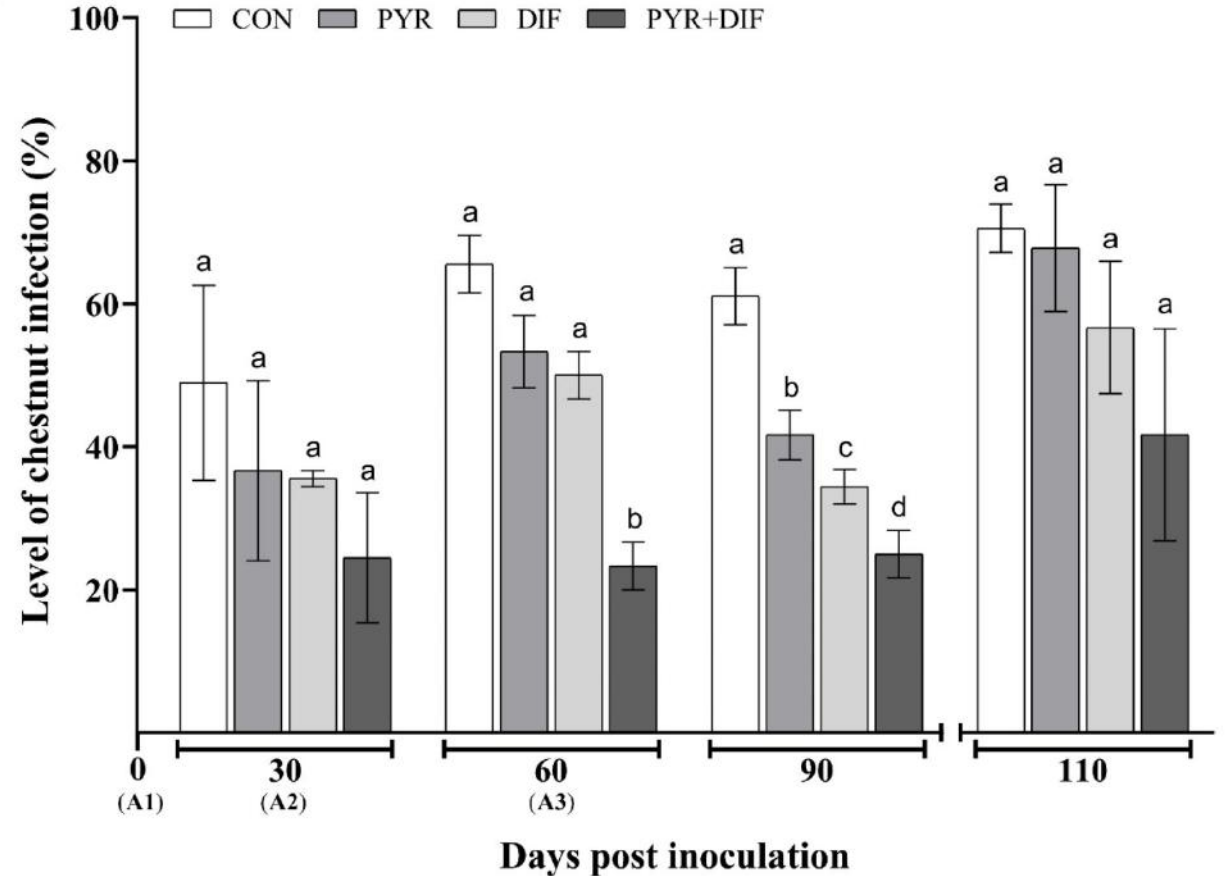
Effect of volatile organic compounds (VOCs) and non-volatile compounds (nVOCs) on mycelial growth of *G. smithogilvyi* isolates B15 and F1N1. **A** Shows the effect of VOCs (Left) and nVOCs (right) on the radial growth of the isolates at each concentration tested (centre). **B** Representation of the effect of nVOCs secreted by the highest three BCAs concentrations on the mycelial growth of both isolates. Plates were incubated at 23 °C in the dark for six days. Means ± SEM labelled with the same letter are not significantly different to the control according to Dunnett's test at $p = 0.05$

Control: Chemical

a



b



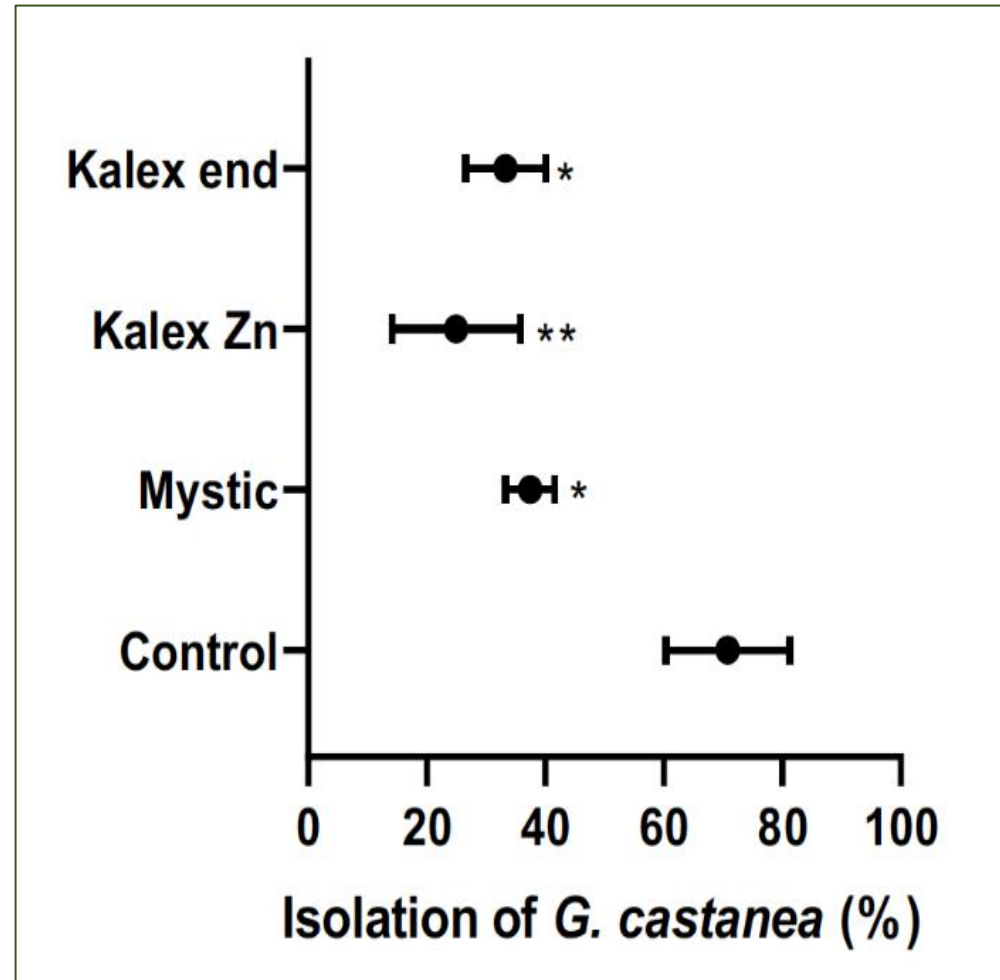
pyraclostrobin (PYR)
difenoconazole (DIF)
their combination (PYR + DIF)

Authors suggest application most effective at flower anthesis

Kalex (Potassium phosphite), Kalex Zn (Zn-phosphonate), and Mystic (tebuconazole)

Year	Product	Application	Rate	Date	Phenological Stage
2019	Kalex [®]	endothecium	0.8 mL/tree	13 June	After bud burst
2019	Kalex Zn [®]	crown spray	3 L/ha	25 June and 6 July	Blooming and burr formation
2019	Mystic [®] 430 SC	crown spray	350 mL/ha	25 June and 6 July	Blooming and burr formation
2019	Control	-	-	-	-
2020	Kalex [®]	endothecium	0.8 mL/tree	23 June	Blooming
2020	Kalex Zn [®]	crown spray	3 L/ha	23 June and 9 July	Blooming
2020	Kalex Zn [®]	crown spray	3 L/ha	23 June; 9 July and 27 August	Blooming; burr development and kernel development
2020	Mystic [®] 430 SC	crown spray	350 mL/ha	23 June and 9 July	Blooming
2020	Mystic [®] 430 SC	crown spray	350 mL/ha	23 June; 9 July and 27 August	Blooming; burr development and kernel development
2020	Control	-	-	-	-

Percent isolation of *Gnomoniopsis smithogilvyi* from chestnut fruits

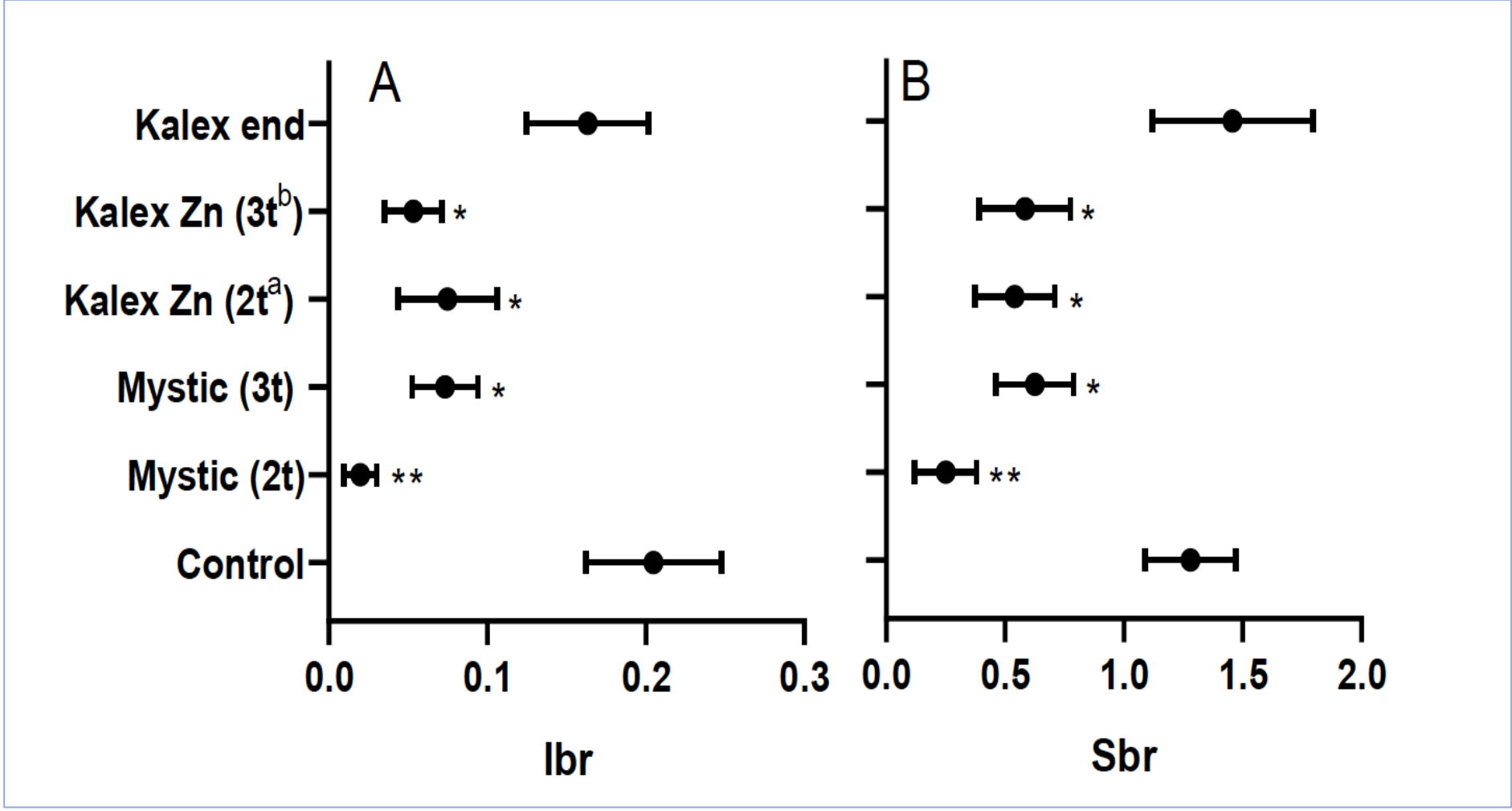


Once, at bud burst

At blooming and burr formation

At blooming and burr formation

Figure reproduced from Bastianelli et al (2022).



Once, at blooming

At blooming, burr de elopement and kernel development

At blooming and burr development, 3 weeks apart

At blooming, burr development, and kernel development

At blooming and burr development, 3 weeks apart

Incidence (A) and severity (B) of brown rot (lbr and Sbr, respectively) of chestnut fruits collected from trees in the six treatment theses in 2020. Bars represent the SEM. Statistical differences with the untreated control are evidenced by * = $p < 0.05$, ** = $p < 0.01$. ^a Two treatments (June and July), ^b Three treatments (June, July and August). Figure and figure caption reproduced from Bastianelli et al (2022b).

Storage and handling

- Infected chestnuts separated from healthy chestnuts by forced air (mechanical harvesting)
- Immersed in water to remove floating debris
- **Curatura:**
 - Hot water (50 C, 45 min)
 - Cold water (10-15 C, 72-96 hours)
- Reduced growth of the fungus immediately after treatment, but not 30 days after.
- Fungistatic rather than fungicidal

Storage and handling

- Adding cell wall degrading enzymes of *Trichoderma* during a hot water treatment (45-50 °C for 50 min; then in 15-18 °C for 50 min) at a ratio of 3:1 (Ruocco et al. 2016).
 - Decreased incidence from 85% to 50% after 2 months.
- Ozone treatment (150 ppb during the day, 300 ppb during the night), at 2.0 ± 0.5 °C; relative humidity at $95 \pm 2.0\%$).
 - After 24 days, nut rot incidence was 25% compared to 87% in control
 - After 5 months, incidence increased to 75%

Future directions

- Integrated control
 - Remove litter to reduce inoculum?
 - Chemical/phosphonate salt applications
- Disease forecasting model – need more info epidemiology and factors associated with disease eg what are the temperature thresholds, how long etc.
- Effect of chemicals on endophyte community