



Transmission of T-2 and HT-2 to beer from raw materials

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Outline of BRI project

- Incubation of barley with a toxigenic *Fusarium* strain
 - to produce artificially high levels of T-2 and HT-2 toxins allowing accurate tracking of the fate of the toxins in processing
- Pilot scale malting of infected barley
 - to determine the fate of the toxins during malting
 - to produce material for pilot brewing
- Pilot scale brewing using infected malt
 - to determine the fate of the toxins during brewing, fermentation and end processing of beer



Production of artificially infected barley

- 50kg barley placed in a rotating drum
- 10 litres *F. sporotrichioides* culture sprayed onto barley
- Barley incubated for 19 days at 20°C
 - Small scale studies showed this to be an adequate period to produce high levels of toxins



Pilot-scale malting with artificially infected barley

Raw barley grain



Steeping



Germination



Kilning



MALT

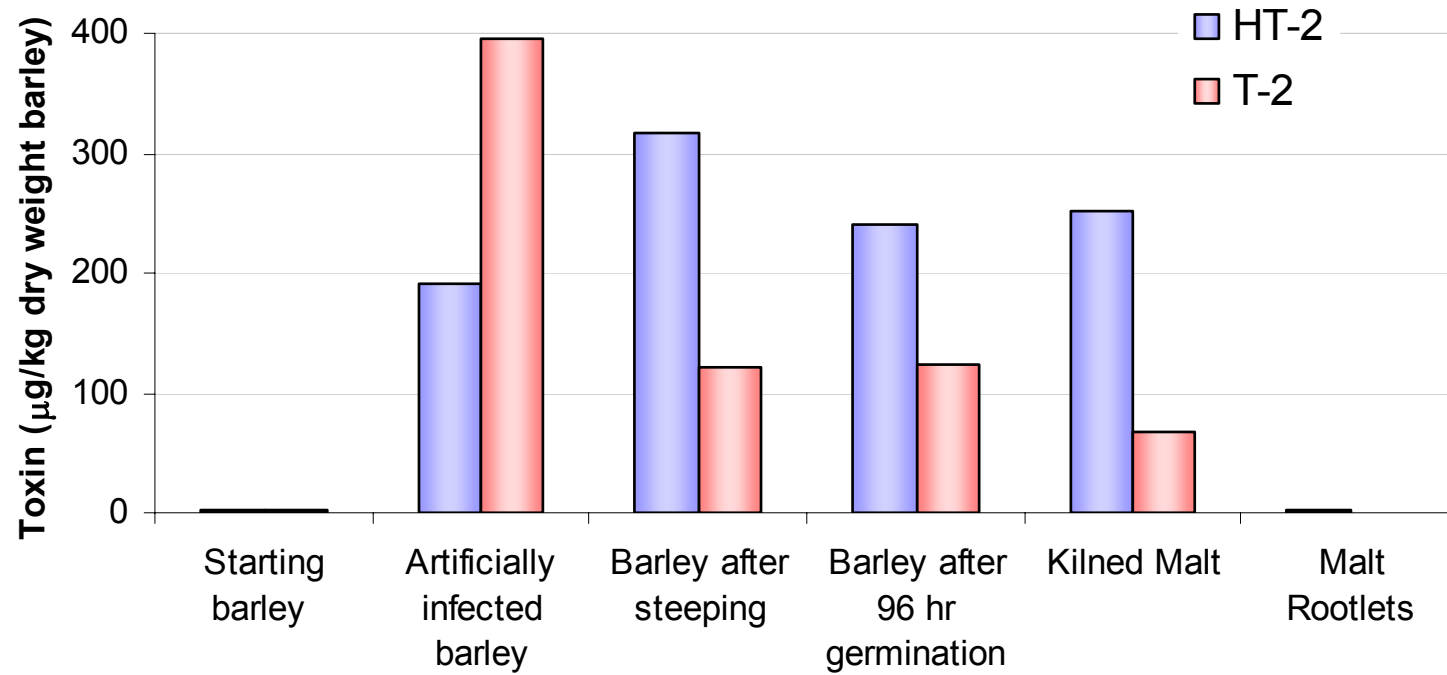


Analysis of T-2 and HT-2 in barley and malt samples

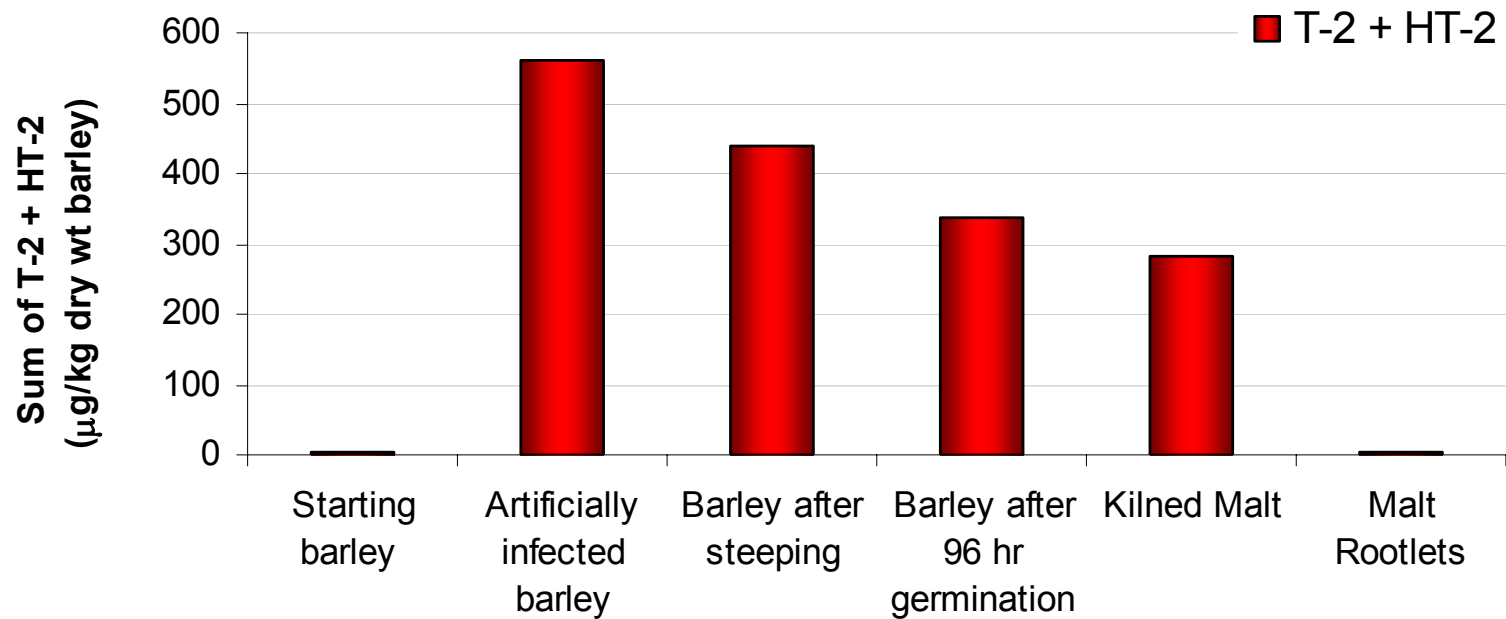
- T-2 and HT-2 were analysed by a validated GC/MS method
- The moisture content of samples varies considerably through the incubation and malting processes
 - therefore results are corrected for moisture content and expressed as μg toxin per kg of barley dry weight



T-2 and HT-2 levels through malting



Combined T-2 + HT-2 levels



Conclusions from malting study

- Overall, there is a significant loss of combined T-2 and HT-2 during malting
 - Most T-2 is lost during steeping
 - Some T-2 may be converted to HT-2
 - Negligible losses with rootlets



Pilot brewing with contaminated barley malt

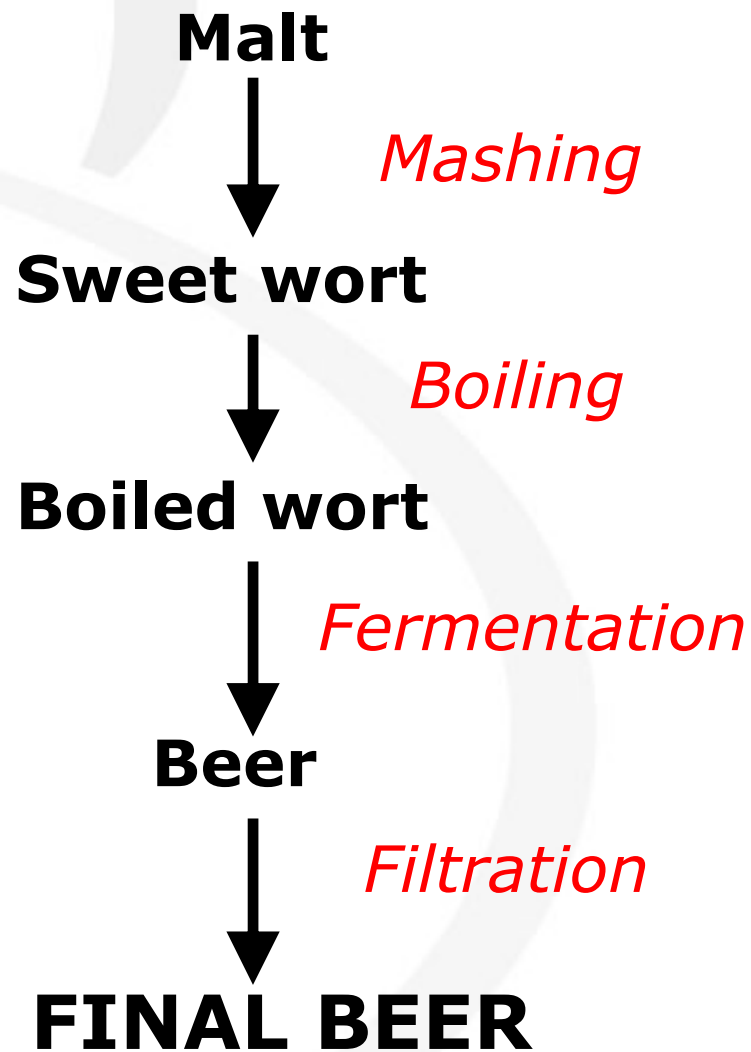


Analysis of T-2 and HT-2 toxins

- Because there is a large dilution during brewing, a more sensitive analysis method must be used for the mycotoxins
- Sensitive and specific LC-MS/MS method developed
 - measures to 1.0 ug/kg in malt and to 0.05 ug/litre in beer for each toxin
- Method validated for malt, beer and intermediate process samples



Brewing Process

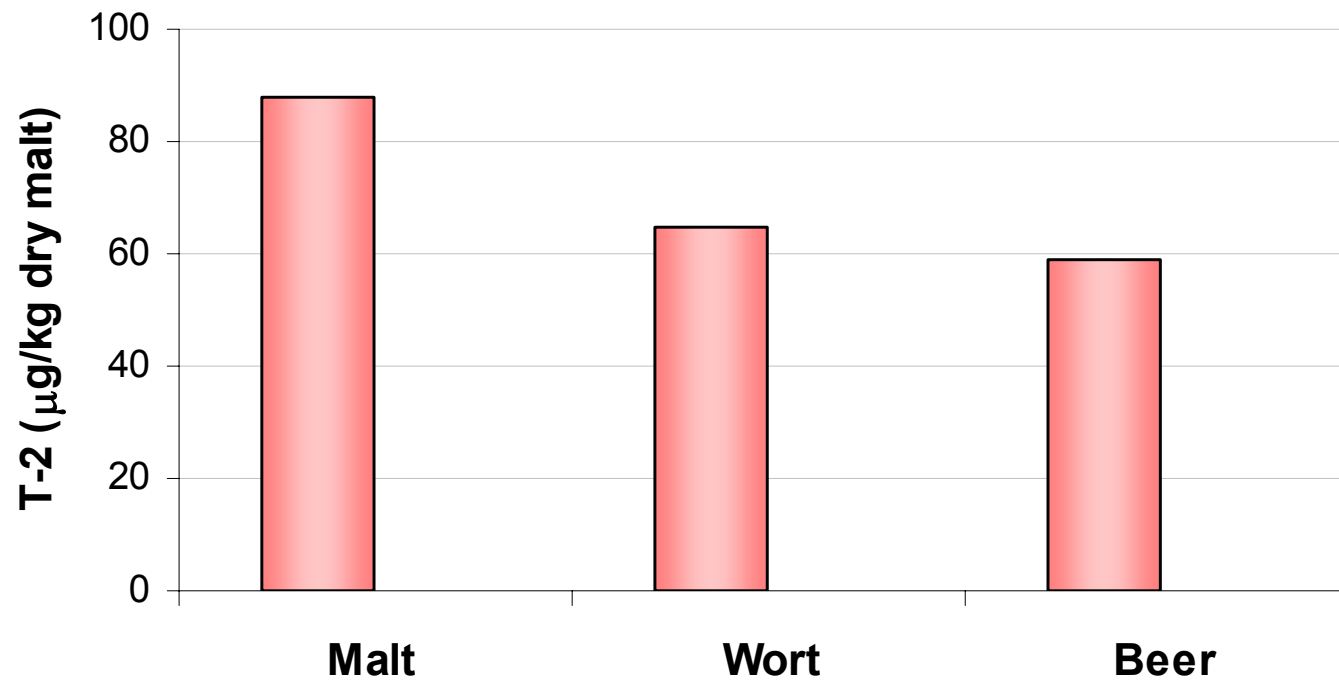


T-2 and HT-2 in brewing fractions

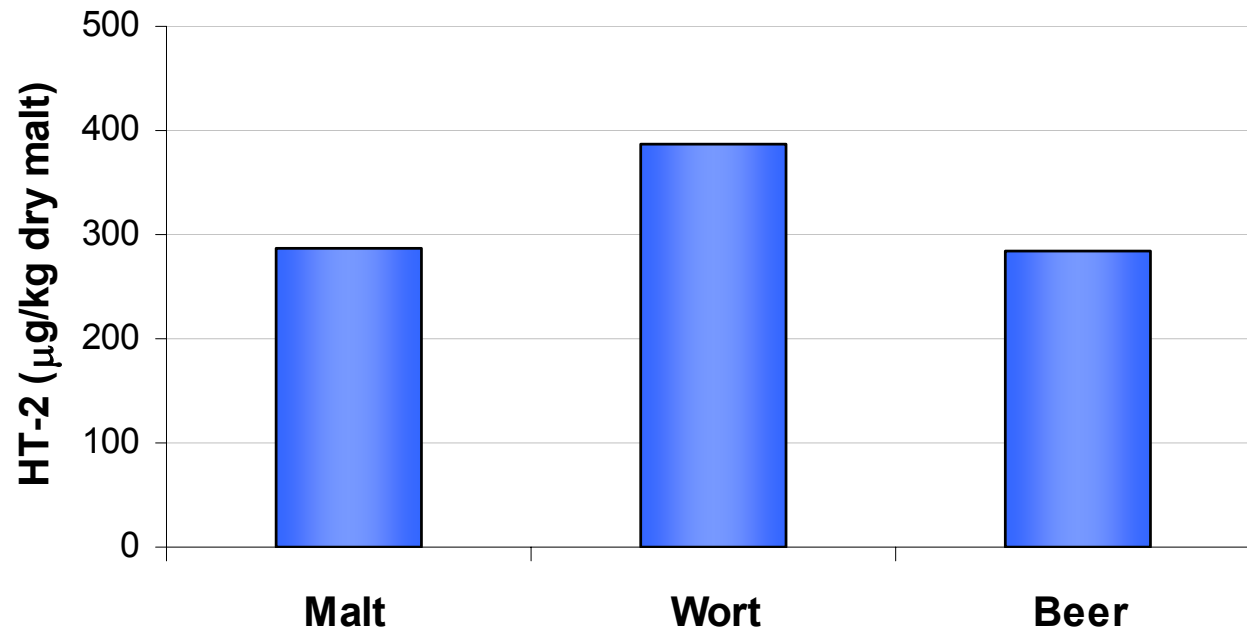
- The brewing process involves dilution and changes of volume
- Toxin concentrations are therefore presented per kg of starting malt to show overall changes during brewing



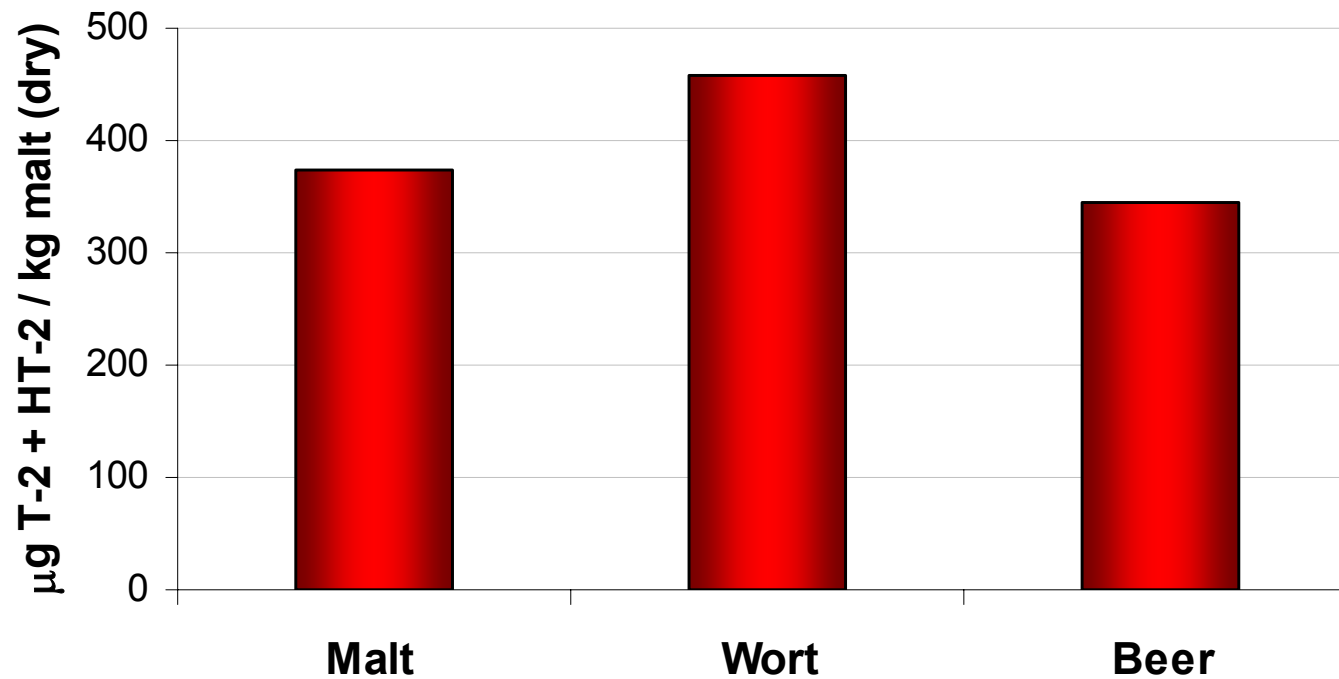
T-2 levels through the brewing process



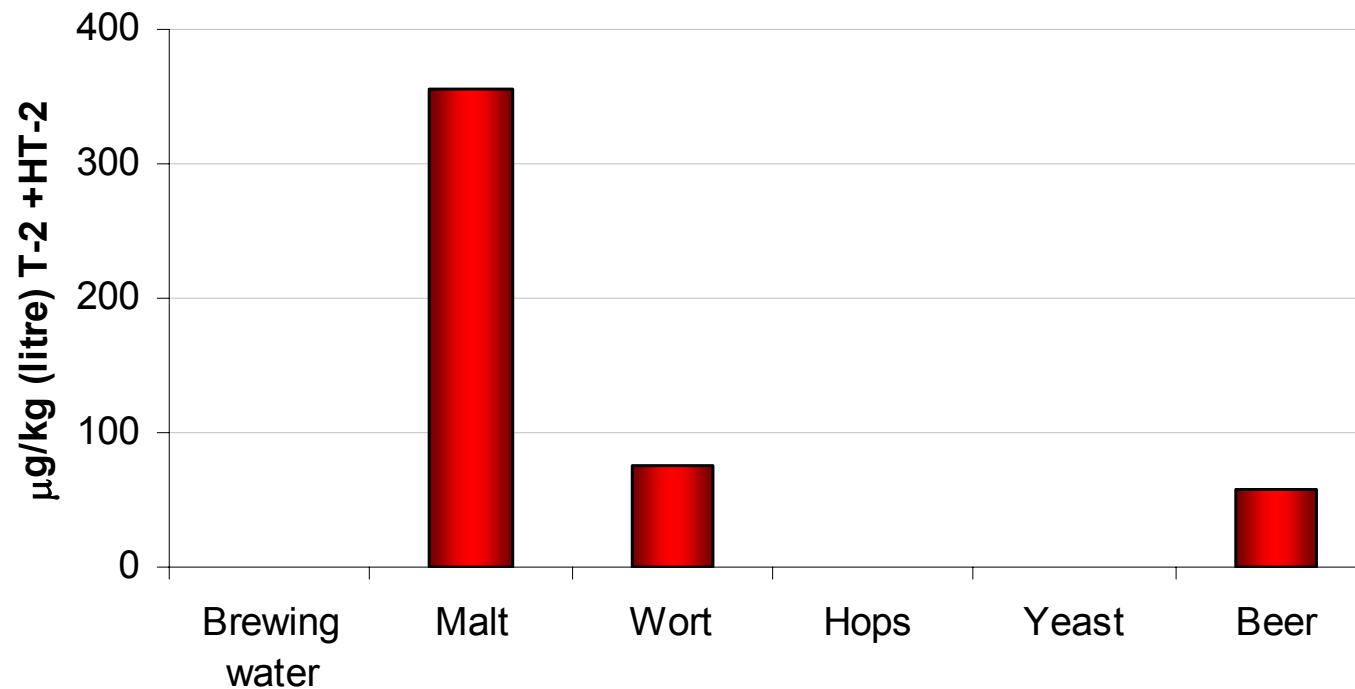
HT-2 levels through the brewing process



Combined T-2 + HT-2 through the brewing process



Actual T-2 + HT-2 concentrations through the brewing process



Conclusions from barley brewing study

- Most of the T-2 and HT-2 in the malt persists into the beer
 - Toxins are completely extracted into the wort during mashing
 - No toxins are derived from the hops, water or yeast
 - No significant loss of toxins with the brewers grains or surplus yeast
- Dilution during brewing reduces the concentrations of T-2 and HT-2 considerably

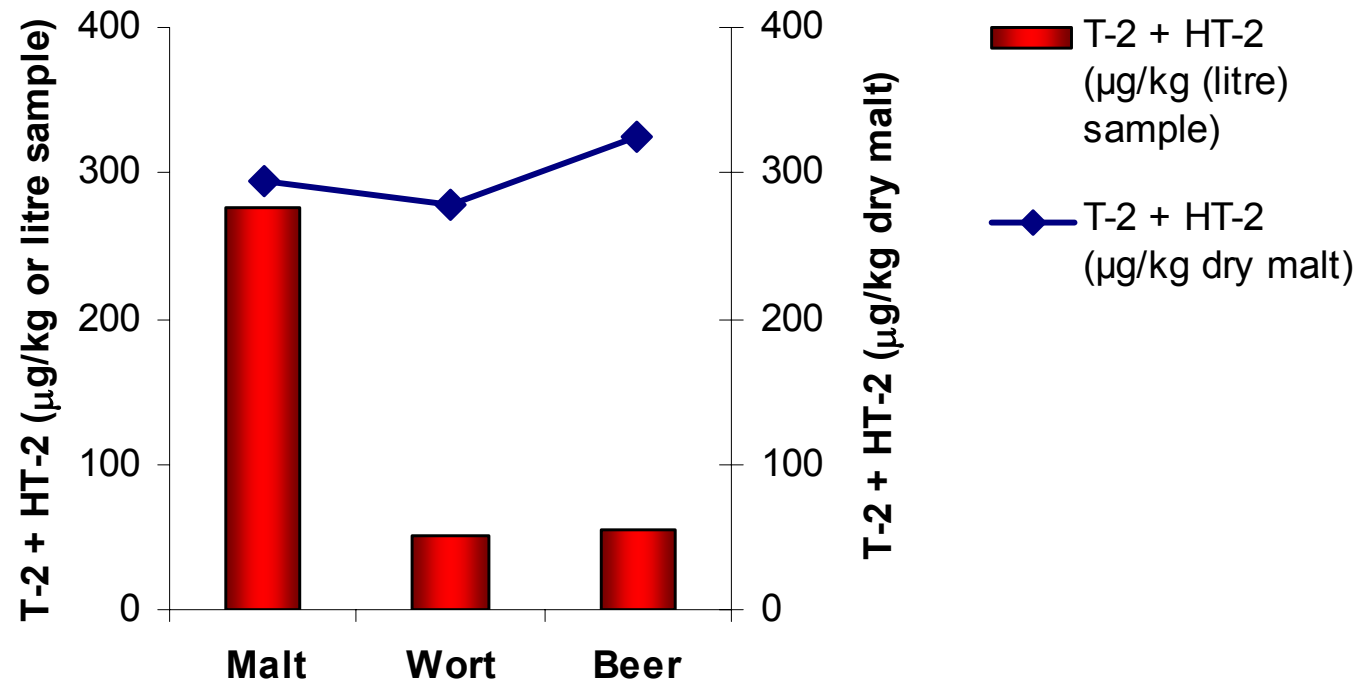


Pilot brewing of wheat beer

- Different style of beer – some of the barley malt is replaced with wheat malt
 - Mashing takes places under different temperature conditions
 - Fermentation step is longer
 - Beer is not filtered
- Therefore potentially more scope for either losses of or increases in T-2 and HT-2



T-2 and HT-2 during brewing of wheat beer



Conclusions from brewing of wheat beer

- Pattern of toxin behaviour very similar to barley malt brew
- All the T-2 and HT-2 in the malt persist into the beer but dilution during brewing leads to much lower final concentrations



Overall conclusions from trials with artificially infected barley

- Significant losses of combined T-2 + HT-2 during malting
- No significant removal of either T-2 or HT-2 in brewing co-products (brewers grains and surplus yeast)
- No significant increase or loss of T-2 or HT-2 during brewing
- Concentrations of T-2 and HT-2 in the final beer are much lower than in the initial malt



Thank you for your attention

