

# Grower Summary

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## **CP 140**

Optimising the use of biocontrol agents to improve the control of botrytis cinerea in key vegetable and fruit crops

Annual 2016

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Before using all pesticides check the approval status and conditions of use.

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**Project title:** Optimising the use of biocontrol agents to improve the control of botrytis cinerea in key vegetable and fruit crops

**Project number:** CP 140

**Project leader:** Prof. Xiangming Xu, NIAB EMR

**Report:** Annual 11/2016

**Previous report:** N/A

**Key staff:** Prof. Xiangming Xu  
Prof. Naresh Magan

**Location of project:** NIAB EMR

**Industry Representative:** Richard Pett, JEPCO, JEPCO (Marketing) Ltd, Norfolk House Farm, Gedney Marsh, Holbeach, Spalding, Lincolnshire PE12 9PB.

**Date project commenced:** 05/10/2015

**Date project completed** 05/10/2018  
**(or expected completion date):**

# GROWER SUMMARY

## Headline

This is the first year of three year-PhD project with much of the time spent on developing experimental protocols. The propidium monoazide quantitative polymerase chain reaction method was developed for quantifying viable populations of *Bacillus subtilis* Strain QST 713 (Serenade) and PreStop (*Gliocladium catenulatum* strain j1446) from crops. The method was developed for advancing the research as no prior method existed for counting the viable numbers for both these biocontrol agents in a sensitive manner. In vitro dose experiments (experiments on agar plates) have suggested that both the BCAs work at suppressing *Botrytis cinerea* colonisation (the fungal pathogen responsible for grey mould) at  $10^8$  colony forming units or spores per droplet. In vivo studies (studies on lettuce plants) have suggested both BCAs significantly reduce disease severity and or stop infection at  $10^8$  colony forming units or spores a ml which is the same as 100L/hectar.

## Background

Biocontrol agents are living organisms and as with every organism reproduction is critical to its survival. Understanding how environmental conditions affect survival, reproduction, dispersal and biocontrol efficacy is crucial to expanding their use. The overall aim of the study is to obtain ecological knowledge on biocontrol agents available in the UK and utilize the knowledge to produce strategies of applying biocontrol agents to improve efficacy against *B. cinerea* on lettuce and strawberry crops.

## Summary

Biocontrol products such as Serenade and PreStop are formed by biological organisms. There were no appropriate ways available on how to estimate the actual numbers of these organisms on the crop after spraying. A large section of the first year focused on forming a methodology in which these organisms can be counted from the leaf surface. Knowing the amount of biocontrol agent product on the crops surface is important since a certain amount is needed to control pathogens such as botrytis the causal agent of grey mould.

Studies on agar plates have shown that a  $10^8$  cfu/droplet (Serenade) and  $10^8$  spores/droplet (PreStop) is needed to suppress the growth of botrytis. Studies on lettuce plants have shown that  $10^8$  cfu/ml of Serenade and  $10^8$  spores/ml of PreStop is needed to stop and or significantly reduce disease severity by botrytis (results will be included in second year annual report).

Now that we know how much of the biocontrol product we need, and have found a way to count/measure the amount of biocontrol agent on a crops surface; we can monitor the effect of the environment on the biocontrol agent product. Finding which environmental conditions effect the biocontrol agent number can be important for understanding when to apply the biocontrol agent product and to improve how well the biocontrol agent product works.

A large portion of the next stage in the research will be focused on finding out which environmental conditions within growing environments across the U.K such as glasshouses and polytunnels may

allow the biocontrol agent product (Serenade and PreStop) to increase in number to better protect the crop.

Preliminary conclusions are drawn only from graphical representation of the data and require further statistical analyses for dose response experiments. We have developed a reliable and robust molecular based assay to quantify *B. subtilis* (Serenade) and *G. catenulatum* (PreStop) viable propagules with the PMA-qPCR technique. The threshold BCA dose for disease suppression, though temperature dependant, is estimated to be  $10^8$  cfu/ml for both Serenade and  $10^8$  spores/ml for PreStop.

The next stage of the research will focus on monitoring the number of biocontrol agents on lettuce and strawberry plants for ten days in different climatic treatments formed by different temperature and humidity combinations. The selected climatic treatments will be representative of temperature and humidity combinations which occur in glasshouse and polytunnel environments all year round, additionally the selected climatic treatments will also be representative of field environment between March to May.

The data collected will be used to form a model capable of estimating the biocontrol agent number up to ten days depending on the climate subjugation after the biocontrol agent has been sprayed onto the crop.

Once the model has been completed research effort will be focused onto how the biocontrol agent disperses on living leaf tissue and if rainfall can effectively disperse biocontrol agents from elder leaves to newly developing leaves.

## **Financial Benefits**

Even though at this stage there are no financial benefits in year 1, forming an understanding on how the climate effects Serenade and PreStop populations may optimise the use of the biocontrol agents which could have financial benefits.

## **Action Points**

- Identify the impact of commercial growing conditions (Temperature and relative humidity combinations) on *B. subtilis* (QST 713) and PreStop viable population's overtime (10 days).
- Identify the colonisation pattern of *B. subtilis* (QST 713) and PreStop on growing leaf tissue.
- Identify the effect of natural rainfall event on *B. subtilis* (QST 713) and PreStop viable dispersion onto newly developing leaves.