## LEF-5830 FISIOLOGIA E BIOQUÍMICA FITOPATOLÓGICA

2º Semestre de 2016 - Aula prática - 23.09.2016

Assunto: Mecanismo estrutural de resistência pós-formado (suberização).

## EXPERIMENT 2. PHYSICAL DEFENSES OF POTATO AGAINST SOFT ROTTING BACTERIA

Plants can defend themselves in many ways when they are wounded or when a pathogen tries to attack. These methods include induced structural and biochemical defenses. In this experiment, potato pieces that have had or have not had time to induce defensive mechanisms will be challenged with the soft rot bacterial pathogen *Pectobacterium* (*Erwinia*) carotovora. Host tissues will be examined for induced defensive mechanisms and how these mechanisms affect infection by the pathogen (M. Windham, personal communication).

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## Procedure 28.2 Physical Defenses of Potatoes Against Soft Rotting Bacteria

- One week before lab, start cultures of P. carotovora in 250-ml flasks containing 75 ml of nutrient broth. Incubate at room temperature on a rotary shaker set for 100 rpm.
- Two days before lab, pour the contents of two flasks into the soil of each flat and mix well.
- One day before lab, take half the potatoes and cut them into pieces that are about 5 cm<sup>2</sup>.
- The day of the experiment, cut the remaining potatoes into pieces that are about 5 cm<sup>2</sup> in size.
- 5. With the scalpel, make thin sections from a potato cut the day before and from a freshly cut piece of potato. Place thin sections of both on a microscope slide with a drop of water and place a cover glass on top of the specimens. Examine the thin sections microscopically and record any differences in the sections.
- 6. Take pieces of freshly cut potato and pieces that were cut the previous day and lay them down (wounded side down) on the surface of the soil. Place flats in the greenhouse for one week. Keep flats moist, but not flooded.
- After a 1 week incubation, remove the potato pieces and examine them for soft rot symptoms. Record your results.

## Anticipated results

The response of the potatoes to wounding can be observed microscopically. In the sections from the potato cut a day prior to the experiment, a white, milky film should be evident. The film should be missing from the freshly sliced potatoes. There should also be differences in disease incidence and disease severity between the 1-day-old pieces of potato and the freshly wounded pieces of potato after they have incubated for I week on the surface of the soil infested with *P. carotovora*.

- Comente sobre o papel da suberização nas interações planta-patógenos;
- Com base no experimento conduzido, que recomendação você faria aos produtores de batata?
- Caso o experimento não tenha funcionado adequadamente, comente sobre as possíveis causas;
- Você conhece algum outro sistema hospedeiro-patógeno, o qual poderia ser usado para a observação de mecanismos estruturais de resistência? Como você conduziria um experimento para ilustrar esse(s) mecanismo(s)?

<u>Literatura</u>: Gwinn, K.D., Greene, S.E., Green, J.F. & Trently, D.J. Host defenses. In: Trigiano, R.N., Windham, M.T. & Windham, A.S. **Plant pathology. Concepts and laboratory exe**rcises. CRC Press, Boca Raton, 2004. p. 261 – 268.