Epidemiology of *P. infestans* in relation to tuber blight. Survival of *P. infestans* sporangia in field soils

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Summary
Infected tubers are a main source of primary inoculum for potato late blight epidemics. To estimate the risk of occurrence of tuber blight “decision support rules” are being developed. Infection of tubers depends on a number of key factors which have to be identified and quantitated. One of those key factors is survival of sporangia in the ridge. Once sporangia have been washed into the ridge they may immediately infect tubers or survive in the soil until conditions become favourable for tuber infection again. Depending on soil climatic conditions, sporangia of *P. infestans* can survive in potato ridges for up to two months. Long term survival of sporangia under field conditions has to be taken into account when developing a strategy to control tuber blight.

Keywords: potato; *Solanum tuberosum*; control strategy;

Introduction
Infected tubers are a main source of (primary) inoculum for late blight epidemics. Prevention of tuber infection during the growing season could be a major contribution to disease control. Formation of sporangia in the foliage and transport of sporangia to the ridge and tubers is considered a prerequisite for tuber infection to occur. Subsequently tubers can be readily infected by *P. infestans*. Sporangia are washed down with rain upon the ridges and into the soil if sporulating *P. infestans* is present in the foliage during rain showers. Especially
tubers near the soil surface were shown to be vulnerable, indicating transport of sporangia and zoospores into the soil by rain water (Lacey, 1966). Once sporangia have been transported into the ridge they can infect potato tubers. Depending on the variety grown infection might occur frequently or sporadically.

The aim of the project is to develop “decision support rules”. These rules can be used for forecasting, to identify risk situations for tuber infection during the growing season and generate a warning accordingly. Also the rules can be used for back casting, meaning retrospective identification of potato lots with a high risk on tuber infection. Weather data and crop growth data are used to identify such lots. In order to do so we had to quantify key factors involved in tuber infection. This paper describes the survival of sporangia in the potato ridge as one of the steps to develop “decision support rules”

**Materials & methods**

Soil samples

Soil was sampled from agricultural land at Wageningen, Lelystad, Valthermond and Vredepeel. These locations differ in soil type and the latter three represent the most important potato growing regions in The Netherlands. No potatoes were grown on the sampled fields for at least two years. Soil samples were stored in polythene bags at 4 °C in the dark until use.

Culturing and inoculum preparation.

Isolate IPO98014 was maintained on potato leaves and tuber slices, cultivar Bintje, in an alternating sequence. One week before the start of the experiments detached potato leaves were sprayed with a sporangial suspension of IPO98014. Leaves were placed on water agar in Petri dishes. These were placed in trays and wrapped in transparent polythene bags. Inoculated leaves were incubated at 15 °C in a climate chamber, with a 16 h light period, during one week. Sporangia suspensions were prepared by rinsing the potato leaves with tap water and adjusting the sporangial concentration in such a way, that for each combination of soil type and soil moisture content 5000 sporangia per gram soil was achieved.
Incubation of the inoculated soil samples
Inoculated soil was put into mesh bags. These bags containing sporangia of *P. infestans* were buried in potato ridges. At each location 4 (replicates) x 15 bags were buried at 5, 10 and 20 cm depth in the ridge.

Survival and viability of *P. infestans* in soil samples
Survival of *P. infestans* was established using the tuber slice test developed by Lacey (1965). Viability of *P. infestans* sporangia was assessed more or less weekly. Sampling dates were adjusted to the expected recovery of *P. infestans*. Sampling was stopped when no viable *P. infestans* could be detected for two sampling dates in a row, or when no more of the 15 samples were available.

Slices of Bintje tubers were cut to a thickness of approximately 0.5 cm and placed individually in a Petri dish. From each of the samples ten times 1 g of soil was put on individual tuber slices. 500 µL of tap water was added onto the soil. The wetted soil was then evenly distributed on the potato slice and the Petri dish was closed. Petri dishes were placed in plastic boxes which were wrapped in transparent polythene bags. The boxes were placed in a climate chamber at 15 °C and a day / night period of 16 / 8 hours. Tuber slices were turned and cut in eight equally sized parts (octants) after 1 day of incubation and placed back in the climate chamber. The number of octants infested with *P. infestans* was established visually after 7 days of incubation. To confirm the presence of *P. infestans*, occasionally mycelial growth was checked for the typical sporangiophores under a light microscope.

Data analysis
The experiments were carried out at four locations with four replicates each time. Statistical analyses were carried out using Genstat (Payne et al, 2002).

**Results**
The recovery of viable sporangia is given in Table 1. The viability lasted longer in 2004 than in 2005. The depth to which the sporangia are buried had little effect upon survival of the sporangia.
Table 1. Final date of recovery of vital P. infestans sporangia (days after inoculation) in the ridge at different depths in fields

<table>
<thead>
<tr>
<th>Location</th>
<th>soil type</th>
<th>Year</th>
<th>Viable sporangia recovered in days after inoculation at different depths</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>5 cm</td>
</tr>
<tr>
<td>Wageningen clay</td>
<td>2004</td>
<td>64</td>
<td>50</td>
</tr>
<tr>
<td>Lelystad clay</td>
<td>2005</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>Valthermond peat-like</td>
<td>2005</td>
<td>35</td>
<td>28</td>
</tr>
<tr>
<td>Vredepeel sand</td>
<td>2005</td>
<td>35</td>
<td>28</td>
</tr>
</tbody>
</table>

Discussion

Survival of P. infestans sporangia under field conditions was still found two months after inoculation. Survival of the sporangia is influenced by many factors of which the strain used is one. IPO98014 is one of the most aggressive isolates in the collection of Plant Research International. A preliminary test with 6 different isolates showed that IPO98014 was one of the two isolates which survived the longest (data not published). For building a model to estimate tuber infection risk or “decision support rules”, data should be based on worst case scenarios, i.e. long term survival of sporangia.

Survival of sporangia is influenced by lysis due to colonisation of sporangia by fungi and bacteria and fungistasis (Andrivon, 1994). Survival of sporangia in 2004 was longer than in 2005. The 2004 season can be characterised as dry, whereas 2005 should be characterised as wet. Survival of sporangia under dry conditions lasts longer than under wet conditions (data not published). Probably under wet conditions sporangia germinate and either infect a potato tuber or die, whereas under dry conditions fungistasis may occurs until conditions become favourable for germination. Also colonisation of sporangia by bacteria and fungi might be more important under wet than under dry conditions.

Maximum persistence of sporangia in a clay soil was 77 days in the UK (Zan, 1962). Survival of P. infestans sporangia up to 45 days was found in France (Andrivon, 1994). These tests were carried out in the laboratory and not under field conditions. Our results were obtained in a field situation showing survival of sporangia in line with other published results (Zan, 1962, Lacey, 1965; Andrivon, 1994).
Long term survival of sporangia might implicate that protection from tuber infection is already needed at flowering. If a potato crop is infested at flowering spores may be washed into the ridge and remain viable until the onset of tuberisation. If conditions are favourable, surviving sporangia might infect newly formed tubers. It is unknown whether this scenario occurs in the actual field situation. Results from our experiments suggest that sporangia at least can survive long enough. The question remains whether the numbers of sporangia surviving are high enough to infect growing potato tubers to some quantity. Nevertheless in agricultural practise it is recommended to protect tubers from as early as the first onset of tuber formation.

Conclusions
Sporangia of *P. infestans* can survive up to 2 months in potato fields. Data generated in the experiment are incorporated into a tuber blight risk model. Tuber protection should start as early as the onset of tuber formation if infection risks of late blight occur.

References