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Inoculation of mature pedunculate oaks (*Quercus robur*) with the root rot fungus *Collybia fusipes*: Relationships with tree vigour and soil factors

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Abstract

The severity of natural infections induced by the root rot fungus *Collybia fusipes* depends on soil factors. Severely infected trees usually show reduced vigour, as evidenced by poor growth. However, it is not known whether reduced vigour could be a cause of the severe infection. The objective was to clarify the relationships between soil factors, tree vigour and susceptibility of *Quercus robur* to *C. fusipes* by artificially inoculating mature trees. Two experiments compared oak trees of different ages and dominance class and oak trees growing on different type of soils. The inoculum survival and the infection success were poor in both experiments when waterlogging was severe. Inoculum survival was better in soil with increasing sand/clay ratio, carbon/nitrogen ratio and decreasing pH and calcium, magnesium and phosphorus availability. The relationships between oak vigour and success of infection or surface area of lesions were inconsistent, with a slightly higher infection success on co-dominant/suppressed oaks in one experiment and a higher surface area of successful infection on dominant oaks in the second.

Introduction

Collybia fusipes causes a root rot on pedunculate oak (*Quercus robur*), sessile oak (*Q. petræa*) and red oak (*Q. rubra*), and on beech, (*Fagus sylvatica*), chestnut (*Castanea sativa*), hornbeam (*Carpinus betulus*) and hazelnut (*Corylus avellana*) (Buller, 1958; Kreisel, 1961; Delatour and Guillaumin, 1984; Guillaumin et al., 1985). This fungus is widespread in the oak forests of north east France where it develops especially at the base of pedunculate and red oaks (Marçais et al., 1998a; Piou et al., 2002). Artificial inoculations showed that *C. fusipes* is a slow primary pathogen capable of infecting vigorous young oak saplings and mature oaks (Marçais and Delatour, 1996; Marçais and Caël, 2000). In field conditions, *C. fusipes* spreads by the way of basidiospores (Marçais et al., 1998b) and natural infections result in root destruction, the severity of which strongly depends on soil factors. Indeed, *C. fusipes* rot is more frequently reported in sandy soils and the likelihood of dieback of an oak with signs of *C. fusipes* infection increases with the sand content of the soil (Camy et al., 2003c). At the stand level, severity of natural *C. fusipes* infections increases with decreasing severity of waterlogging. (Piou et al., 2002, Camy et al., 2003a).

Trees infected with *C. fusipes* have poor growth (Marçais and Caël, 2001). However, because root rots are slowly developing diseases and growth reductions are usually long term, it is difficult to determine to which degree the pathogen is the cause of the poor tree vigour, by which we mean a growth slower than the neighbour trees for a long period. Alternatively, trees with poor vigour could show an increased susceptibility to *C. fusipes* and be preferentially infected.

The objective was to clarify, under field conditions, the relationships between soil factors, tree vigour and the susceptibility of *Q. robur* to *C. fusipes* in the early stages of the infection process.

Materials and methods

Fungus isolates and inoculum production

Four isolates of *C. fusipes* were used. C49 was isolated from a red oak in 1992 at Les Barres, Loiret, C41 and C52 were isolated from pedunculate oaks in 1994 at Siarrouy, Haute-Pyrénées and in 1993 at Mersuay, Haute-Saône respectively. C62 was isolated from sessile oak in 1994 at Amance, Meurthe-et-Moselle.

Pieces of wood were colonized by *C. fusipes* (Marçais and Delatour, 1996). Briefly, stems of hazel, *C. avellana*, 1.5–2.5 cm in diameter, were collected and cut into segments 3 cm long. They were placed in glass jars filled with tap water and sterilized twice at 120 °C for 30 min, 24 h apart. The water was drained at the end of each sterilisation. A liquid malt medium was added to cover half the height of the wood segments and a third sterilisation was done for 20 min at 120 °C. To improve aeration, a hole was drilled in the jar top and plugged with cotton wool. Each of the jars was aseptically seeded with blocks of inoculum ($0.5 \times 0.5\,cm^2$) from a *C. fusipes* culture on malt agar ($20\,g\,l^{-1}$ malt Difco, $15\,g\,l^{-1}$ agar) and incubated for 30–45 days at 23 °C. All the liquid was drained from the jars with a syringe and they were further incubated at 23 °C for 8–9 months.

Inoculation experiment I

Sixty mature pedunculate oaks were selected for inoculation in 17 stands from the forest of Amance, Meurthe-et-Moselle. The stands were selected to represent different tree ages (50–200 years old) and because of the presence of *C. fusipes* root rot. They were mixed pedunculate and sessile oak stands with an understory dominated by hornbeam. An effort was made to select stands that were homogeneous as far as vegetation type was concerned. Soils were of a loamy clay texture, with pH of the surface soil in water being 4.9–5.9. The first traces of waterlogging appeared from 5–55 cm under soil level depending on the stands. Sampling was designed to obtain 9 to 12 trees in each of six age classes, about 50, 70–90, 100–120, 120–140, 140–160 and over 160 years old. Two to five trees were selected per stand, with no more than three oaks in a particular age class from the same stand. Thirty-six trees were dominant and the others were co-dominant or suppressed. Care was taken that the suppressed/co-dominant trees were well distributed within the six age classes. Pedunculate oaks were identified with binoculars using leaf morphology and acorns (when present). A wood core to the centre of the stem was extracted at breast height on each tree and was used to estimate its age and its radial growth during the last 10 years. Radial growth was standardized (Becker et al., 1994) to remove the effect of age. Tree height and the tree diameter at breast height were measured. The ratio height/diameter was calculated to reflect the average tree vigour in the past. Severity of waterlogging was estimated for each tree by extracting a soil core at about one metre from the collar to determine the depth of appearance of first signs of mottling, i.e, first reddish traces of insoluble oxidized iron deposition and/or discoloration indicating iron depletion.

In May 1997, the sixty trees were inoculated with the colonized hazel stem segments. Collar roots were exposed and three roots, 2–5 cm in diameter at a depth varying between 10–30 cm, were selected for inoculation. The surface of the bark was brushed and washed with water and a hazel stem segment colonized with C41, C49 or C52 was fastened firmly in contact with the unwounded bark on one of the three selected roots. Each tree was inoculated with the three isolates. A total of 180 roots were inoculated. Six additional control inoculations had uncolonized hazel stem segments attached to the roots. Root diameters were recorded and the soil was replaced. In December 1999, the inoculated roots were excavated and a section of the root with all the infection present was cut and brought back to the laboratory for examination. Survival of *C. fusipes* in the inoculum was assumed when black crusts covered the segment surface and white mycelium was present underneath and/or when the wood had a bright orange colour. Infections often occurred in non-coalescent lesions under and/or in the vicinity of inoculum. The maximum length and width of each lesion in the bark was recorded as well as its depth of penetration within the bark. The global surface area of the lesions was estimated as the sum of geometric means of the two diameters of each surface area $\Sigma(\pi \times (\text{length} \times \text{width})/4)$. Isolation of *C. fusipes* was attempted from pieces of necrotic root bark and wood putatively colonized by the fungus as well as from the inoculum segment. They were washed under water, surface sterilized for 1 min in sodium hypochlorite at 3.75% active chlorine and rinsed 3 times in sterile water. The outer bark was removed and chips of dead bark or decayed wood were plated on MAT medium ($10\,g\,l^{-1}$ of malt Difco, $100\,mg\,l^{-1}$ penicillin, $100\,mg\,l^{-1}$ streptomycin, $250\,mg\,l^{-1}$ thiabendazole and $15\,g\,l^{-1}$ agar). Isolates

that were recovered were paired on malt agar medium with a control isolate of C41, C49 and C52 to check whether they were the ones that had been inoculated.

Inoculation experiment II

Ten plots, where *C. fusipes* was present, were selected in north-eastern France to obtain a range of soil sand content (Table 1). The majority of the plots were pedunculate oak stands with an understory dominated by hornbeam. In the plots of Amance, Adelans, Filain and Quers pedunculate oaks were mixed with sessile oaks. Fifteen mature pedunculate oaks (over 90 years old) were inoculated in each plot. Oak species was determined from leaf morphology. Seventy percent of the trees were dominant and 30% were co-dominant or suppressed. Height and diameter at breast height were measured for each tree and the ratio height/diameter was computed.

In autumn 1998, each pedunculate oak was inoculated with the three isolates of *C. fusipes* C41, C49 and C62. In total, 450 roots (10 plots × 15 trees × 3 isolates) were inoculated. Three additional roots per plot were inoculated with a hazel segment which had not been colonized by *C. fusipes* as controls. Inoculations were excavated 2.5 years later. Isolates that were successfully recovered from the lesions were paired with control isolates of C41, C49 and C62. On each plot, 3 soil samples of 0.5 dm^3 were collected from the first 10 cm, with an earth auger and analysed for texture, organic carbon content (sulfochromic method), total nitrogen content (Kjeldahl method), pH (10 g of soil in 50 ml of deionized water), assimilated phosphorus (Duchaufour method) and exchangeable cations (cobaltihexamin mehod) such as calcium, magnesium, potassium, manganese. All these soil analyses were carried out following the AFNOR norms concerning soil quality (AFNOR, 1999). The bulk density of soil was also evaluated from 15 soil samples per plot. Soil samples of 0.7 dm^3 were collected from the first 15 cm, at the base of each tree, dried at 110 °C for 48 h and weighed. The bulk density was computed as the weight per unit volume. The severity of waterlogging was evaluated at the base of each tree as described in experiment I.

Data analysis

Inoculum survival was computed from the recorded inoculum condition. For the roots where inoculum could not be retrieved (14 in experiment I and 55 in experiment II), it was assumed that survival had occurred whenever a lesion was present on the inoculated root. A lesion was assumed to be present whenever a necrosis with a depth of penetration in the bark over 3mm was present. It is very difficult to determine whether less developed necrosis are induced by *C. fusipes* as their appearance is non-specific and isolation of the pathogen from them is seldom successful. The infection success was computed as the frequency of lesion per effectively inoculated roots, i.e. roots with an inoculum alive. The relationships between inoculum survival, *C. fusipes* isolates and soil factors and between the success of infection, soil factors and tree characteristics were analysed by generalized linear

Table 1. Characteristics of the 10 plots selected for experiment II

Plot	Sand content (%)	Sand/ clay ratio	pH	Carbon/ nitrogen ratio	Waterlogging depth (cm)	Inoculum survival (%)	Infection success (%)
Arpenans	3.4	0.1	4.9	13.5	27	62	74
Filain	7.2	0.2	5.8	13.3	25	77	86
Amance	11.5	0.4	5.7	12.3	29	79	79
Adelans	15.3	0.7	4.3	15.9	63	73	54
Ainvelle P21	26.1	1.2	5.6	13.0	40	69	88
Mersuay	30.3	1.1	5.2	12.9	38	70	85
Quers	33.4	1.7	4.4	15.7	36	78	90
Equevilley	35.0	2.0	4.1	17.1	87	88	94
Ainvelle P28	45.0	1.8	4.5	15.4	40	80	76
Les Aynans	53.0	2.7	4.1	14.6	42	88	67

Note: The sand and clay content was evaluated in the upper most 10 cm of the soil. The pH at the soil surface was measured from 10 g of dry soil in 50 ml of deionized water. Organic carbon (sulfocromic method) and total nitrogen (Kjeldahl method) were measured from soil samples in the first 10 cm under soil surface. The waterlogging depth was the depth at which any signs of iron depletion or deposition occurred.

analysis, using the procedure genmod of SAS (SAS 1989). A binomial distribution of the data was assumed and the logistic link function was used. The model validity was checked using the deviance degree of freedom ratio, by plotting deviance residuals against the linear predictor and with a half normal plot (Collett, 1991). To study the relationship between the surface area of lesion induced by *C. fusipes*, soil factors and tree characteristics, the lesion surface area was log transformed to obtain a normal distribution of residuals and subjected to an analysis of variance using the procedure glm of SAS.

In experiments I and II, we checked that there was no statistical link between tree characteristics, such as crown health status or age, and the inoculum survival before analysing further results.

In experiment II, two types of analyses were performed for variables measured at the plot level (soil factors except depth to waterlogging traces) and at the tree level (depth to waterlogging traces, tree characteristics such as dominance class). For tree level analyses, the plot factor was taken into account by treating plots as blocks. The depth to waterlogging signs was tested in the model at the tree level because waterlogging severity was very variable within most plots. Also traces of iron deposition or depletion in water saturated soil depends on the pH of the soil.

Results

Experiment I

Seven roots could not be retrieved or were infected by a *C. fusipes* isolate that proved to be different from the one inoculated. Thus 173 of the inoculated roots could be analysed. *C. fusipes* was alive in 65% of the hazel stem segments used as inocula and was isolated from all the active segments. Inoculum survival was not significantly different for the 3 isolates (53%, 69% and 74% for C41, C49 and C52, Table 2) and was significantly related to the severity of waterlogging (Table 2). *Collybia fusipes* did not persist as well on the stem segments in the more severe waterlogging conditions, where traces of waterlogging appeared close to the soil surface (Figure 1).

Lesions were present on 54% of the roots and *C. fusipes* was isolated from 98% of successful infections. Eighty-nine percent of successful infections had reached the cambium. Neither tree age nor the isolate influenced infection success, i.e. the likelihood of infection when the pathogen had persisted on the segment (Table 2). Infection successes were 71%, 78% and 91% for C41, C49 and C52. Infection success increased significantly with decreasing waterlogging and was slightly higher on suppressed/co-dominant trees (87 %) compared to the dominant trees (76%, Table 3, Figure 1). The surface area of a successful infections did not depend on tree age (Table 3). However, there was a non-significant tendency for trees of 50 years old to have smaller lesions ($12.0 \pm 1.4\,cm^2$ compared to 16.3–18.9 cm^2 for trees 80 to over 160 years old). The surface area of successful infection was also not affected by waterlogging or tree dominance class and was $14.3 \pm 2.5\,cm^2$ for dominant trees and $12.5 \pm 2.5\,cm^2$ for suppressed/co-dominant trees (Table 3, Figure 2). Two additional measures of tree vigour were used to test the relationship between tree susceptibility to the pathogen and tree vigour, tree radial growth in the last 10 years and the height/diameter of trunk ratio. These two measures were not related either with the infection success or with the successful lesion surface area (results not shown).

Table 2. Factors affecting survival of C. fusipes on hazel stem segments and infection success on mature pedunculate oaks (experiment I): component likelihood χ^2 value associated with the introduction of each effect into the generalized linear model

Source	Survival on hazel stem segments		Infection success[a] χ^2(p value)
	df	χ^2(p value)	
Isolate	2	5.7 (0.059)	4.5 (0.104)
Waterlogging	1	5.0 (0.025)	4.7 (0.031)
Dominance class	1	—	6.5 (0.011)
Tree age	1	—	2.0 (0.161)
Age × dominance class	1	—	1.1 (0.287)

[a]Presence of lesion taking into account only inoculated roots where *C. fusipes* was still present on the hazel stem segment at the end of the experiment.

Experiment II

Because ten trees were wind thrown during the storm of December 1999, 30 roots could not be re-located. Eleven roots had lesions that yielded a *C. fusipes* isolate that was not the one inoculated. So, the results from only 409 of the 450 inoculated roots could be analysed. *C. fusipes* was still alive in 76% of hazel segments and was reisolated from 52% of them. Inoculum

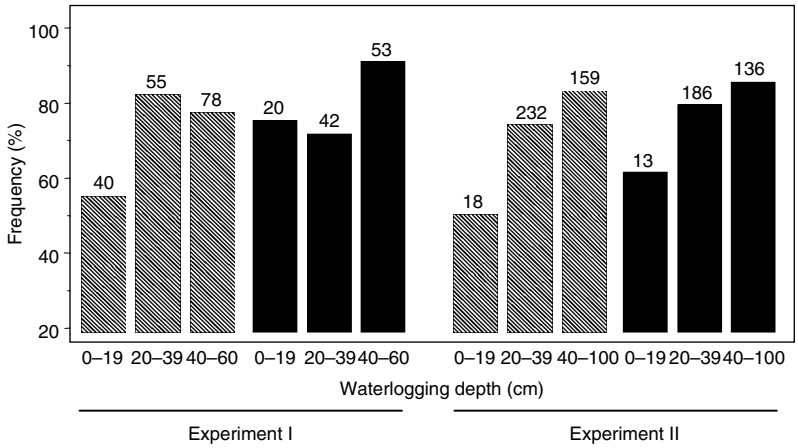

Figure 1. Survival of *C. fusipes* on hazel stem segments (▒) and infection success (■) for inoculations with *C. fusipes* persisting on the stem segment on mature pedunculate oaks (experiments I and II) among different categories of waterlogging severity (depth to first traces of iron deposition or depletion). The number of hazel stem segments or the number of seedlings with active inoculum, in each category of waterlogging, is given above the bars.

Table 3. Factors affecting the lesion surface area of successful infections on mature pedunculate oaks (experiment I): an analysis of variance

Source	df	Mean square	*F* value	*p* value
Model	6	1.04	1.62	0.145
Error	86	0.63		
Isolate	2	0.52	0.41	0.666
Waterlogging	1	0.65	1.03	0.312
Tree age	1	2.39	3.80	0.055
Dominance class	1	0.64	1.01	0.312
Age × dominance class	1	1.04	1.65	0.202

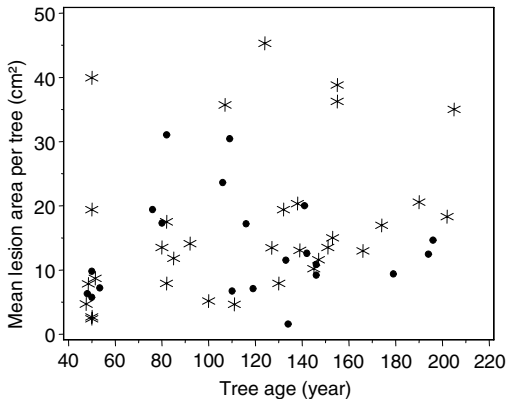

Figure 2. Susceptibility to *Collybia fusipes* of mature pedunculate oaks of different age and competition status (experiment I). ∗, Dominant trees; •, Codominant/suppressed trees. Only successful infections are taken into account.

survival was not different between the *C. fusipes* isolates (Table 4) being 76%, 72% and 81% for C41, C49 and C62. It was not significantly linked with phosphorus, manganese or bulk density (Table 5). By contrast, inoculum survival increased significantly with increasing sand/clay and carbon/nitrogen ratio and decreasing pH, exchangeable potassium, calcium and magnesium content (Table 5, Figure 3). All the soil properties are tightly correlated, in particular pH with exchangeable content, sand/clay and carbon/nitrogen ratio (results not shown). The survival of inoculum was not significantly different in the different plots although it varied from 61% at Arpenans to 88% at les Aynans and Equevilley (Table 4). Within plots, inoculum survival increased with increasing depth to signs of waterlogging (Table 4, Figure 1).

Sixty-six percent of the roots were infected with *C. fusipes*. The rate of *C. fusipes* isolation from the successful infections was only 44%. However, lesions were typical of the type induced by *C. fusipes* as reported in the literature (Marçais and Caël, 2000) and were kept as successful infections. It was assumed that these infections were induced by the inoculated isolates, because infections induced by wild isolates were usually much bigger than those induced by the inoculated ones. Lesions had reached the cambium in 88% of the successful infections. The infection success was not significantly different for the three *C. fusipes* isolates (Table 4) and was 78% for C49 and C62 and 84% for C41. It ranged from 54% in the plot of Adelans to 94% in the plot of Equevilley (Table 1). Infection success

Table 4. Factors affecting inoculum survival, infection success and lesion surface area of successful infections (experiment II, analyses at the tree level)

	Inoculum survival		Infection success		Surface area of lesion	
	df	χ^2(*p* value)	df	χ^2 (*p* value)	df	*F* (*p* value)
Plot	9	11.41 (0.249)	9	11.31 (0.255)	9	1.08 (0.183)
Isolate	2	3.41 (0.182)	2	2.45 (0.294)	2	1.71 (0.183)
Waterlogging	1	4.91 (0.027)	1	8.45 (0.004)	1	0.10 (0.753)
Dominance class	—		1	0.19 (0.660)	1	5.99 (0.015)

Note: Regressors were introduced together in the model, after taking into account the effect of plot. Waterlogging severity and tree characteristics were assessed for every tree.

Table 5. Soil factors affecting inoculum survival, infection success and lesion surface area of successful infections (experiment II, analyses at the plot level)

	Inoculum survival		Infection success		Surface area of lesion	
	df	χ^2(*p* value)	df	χ^2 (*p* value)	df	*F* (*p* value)
Sand/clay	1	6.41 (0.011)	1	0.13 (0.718)	1	0.05 (0.823)
Bulk density	1	3.35 (0.067)	1	0.09 (0.771)	1	0.04 (0.833)
Carbon/nitrogen	1	6.03 (0.014)	1	0.04 (0.834)	1	0.27 (0.605)
pH	1	6.83 (0.009)	1	0.81 (0.367)	1	0.23 (0.633)
P_2O_5	1	1.60 (0.206)	1	2.46 (0.117)	1	0.21 (0.644)
K	1	5.63 (0.018)	1	0.00 (0.999)	1	0.02 (0.887)
Ca	1	4.84 (0.028)	1	1.04 (0.308)	1	0.24 (0.627)
Mg	1	6.40 (0.011)	1	1.35 (0.245)	1	2.50 (0.115)
Mn	1	0.67 (0.412)	1	0.00 (0.975)	1	1.83 (0.177)

Note: Each soil factor was introduced separately in the model. Soil factors were assessed at the plot scale.

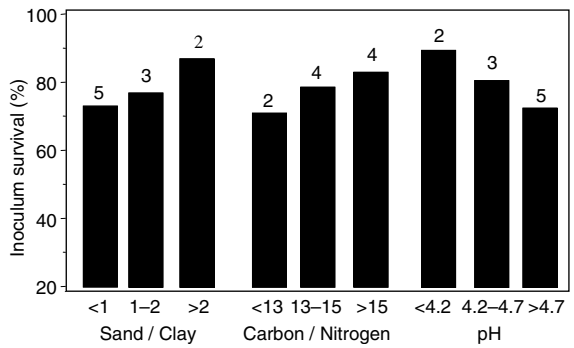

Figure 3. Percentage of *C. fusipes* inoculum survival in soils of different sand/clay ratio, carbon/nitrogen ratio and the soil pH (experiment II). These soil factors were measured at the plot scale. They have been categorized. The number of plots in each category is given above the bars.

was significantly influenced by waterlogging. Indeed it increased with the depth of waterlogging signs (Table 4, Figure 1). It was not significantly linked with any of the other soil factors or tree characteristics (Tables 4 and 5).

C. fusipes isolates did not differ in the size of the lesions they induced (Table 4). The surface area of lesions was not significantly different among plots, although it varied from $30.9 \pm 3.6\,cm^2$ in Mersuay to $54.6 \pm 8.7\,cm^2$ in Filain. Lesion surface area was not linked with any of the soil factors (Tables 4 and 5). By contrast, lesion surface area was significantly linked with the dominance class of the trees and was $33.4 \pm 3.6\,cm^2$ for suppressed/co-dominant trees and of $43.8 \pm 3.6\,cm^2$ for dominant ones (Table 4). As found in experiment I, the ratio height/diameter of trunk was not significantly related either with likelihood of infection success or with successful lesion surface area (results not shown).

Discussion

The early stages of the *C. fusipes* infection process were strongly influenced by soil factors. The pathogen itself and in particular the inoculum survival was clearly affected by the severity of waterlogging and by soil

factors linked to texture such as pH, exchangeable cation availability and nitrogen/carbon ratio. Conversely, there were no effects of tree age on infection success or size of successful infections. Also the influence of the tree dominance class on these two measures of tree susceptibility to *C. fusipes* was inconsistent between the experiments.

The infection success and lesion mean surface areas obtained on mature pedunculate oaks in the two experiments were similar to those already obtained in previous inoculation experiments on mature oaks, *Q. petraea*, *Q. robur* and *Q. rubra* (Marçais and Caël, 2000). In particular, they were very close to what was obtained on red oak, with infection success over 70%. In contrast, infection success on sessile oak was only 33% in this previous work. This could reflect inherently higher susceptibility to *C. fusipes* of *Q. robur* and *Q. rubra* compared to *Q. petraea* and may explain why the disease is more frequent in those two oak species (Piou et al., 2002).

Despite the 2.5 year duration of the experiment, only the early stage of the infection process was studied. Indeed, by field investigations, it was determined that the average period required by *C. fusipes* to affect severely the root system of an oak was about 30 years after the initial infection (Camy and Marçais, 2002). This may be a major pitfall in inoculation studies of slowly evolving root rot pathogen (Redfern, 1983; 1998; Goheen and Hansen, 1994). Indeed, Redfern (1983; 1998) found evidence in a field inoculation experiment with *H. annosum* on Sitka spruce, that whereas infection success was equal in mineral and peat soil after 5 years, large differences in disease development between the two soil types could be evidenced after 10 years. However, in this case, *H. annosum* was introduced via stump inoculation, so that it clearly took a considerable time to spread to the root systems of neighbouring trees. Even in favourable soil conditions, this must have required a longer period than in our case where *C. fusipes* inoculum was directly fastened to the inoculated roots. It is worth noting that short incubation periods (respectively of about 4 and 1 year) were long enough to demonstrate differences in susceptibility to *C. fusipes* of different oak species (Marçais and Caël, 2000) and differences in susceptibility to *Armillaria* of *Q. robur* or *Pinus sylvestris* of different dominance class (Redfern, 1978; Davidson and Rishbeth, 1988).

Soil factors related to survival of inoculum were tightly correlated. *C. fusipes* survived better on woody inocula in coarser and poorer soils. This might be part of the reason why *C. fusipes* is found more in coarse textured soils and why infection has a higher negative impact on tree health in soil with a sandy texture and low pH (Camy et al., 2003c). Blenis et al. (1989) similarly observed a greater survival of *A. ostoyae* inoculum in sandy soils and supposed that such results could be related to the high oxygen availability in such type of soils. This hypothesis could also be applied to *C. fusipes* which appears to be intolerant to hypoxia (Camy et al., 2003b). The negative influence of waterlogging on inoculum survival could also be linked to a strong reduction of oxygen, especially in heavy soils (Lévy and Lefèvre, 2001; Nisbet et al., 1989). Indeed, inoculum survival and infection success decreased in the present study when waterlogging traces appeared close to the soil surface. These results support field observations in which natural *C. fusipes* infection was shown to be more severe when soil is less affected by waterlogging (Piou et al., 2002; Camy and Marçais, 2002; Camy et al., 2003a). However, in the absence of any relationship between soil apparent density and inoculum survival or infection success there remains no evidence that soil texture affects the pathogen directly through oxygen availability. The coarse soils differ from the finer-textured ones for many factors and it is difficult to pinpoint the reason that makes them more favourable to *C. fusipes* survival.

There was no evidence that oak trees growing in the coarser and more acidic soils showed a higher susceptibility to *C. fusipes*, as far as infection success or successful lesion surface area were concerned. This is in contrast with what is reported for other root rots in the literature. Indeed Singh (1983) and Redfern (1978) reported that seedlings of several species of conifers, inoculated with *Armillaria* spp., showed an increased susceptibility when grown in more coarse and acidic soils. Piri (2000) reported a lower survival of the pathogen in roots of Scots pine inoculated with *H. annosum* on plots fertilized 3 years before inoculation, as well as a lower infection success and extension of this pathogen in roots.

Despite the different parameters assessed reflecting tree vigour, such as dominance class, ratio height/diameter or tree growth, there were no consistent relationships between tree vigour and infection success or size of successful infections. This is in agreement with the results of a six year survey of the evolution of *C. fusipes* natural infections on red oaks which did not show correlation between tree vigour and occurrence of new infections or development of established infection (Camy and Marçais, 2002). Results

reported with other root rot diseases are variable. Goheen and Hansen (1994), working on western Hemlock and Douglas fir inoculated with *Phellinus weirii* did not find any correlation between dominance class and infection success or root colonisation. However, Davidson and Rishbeth (1988) demonstrated that *A. gallica*, considered as a secondary pathogen, was able to infect only suppressed trees in field inoculations, whereas *A. mellea* and *A. ostoyae,* considered as primary pathogens, could attack co-dominant trees. Also Redfern (1978) showed that suppressed Scots pines inoculated with *Armillaria* spp were more susceptible than dominant ones. Tree past vigour might not be representative of its actual vitality which could be defined as its capacity to survive in a next future, and can often be characterized by amount of energy reserves, such as root starch content (Wargo and Houston, 1974). However we did not quantify any reserves and can therefore draw no conclusions on the relationship between susceptibility to *C. fusipes* and tree vitality.

Quercus robur is a species requiring good availability of water and mineral nutrients (Becker and Lévy, 1990). Soils favourable to *C. fusipes* often have a deficiency of these requirements, which could lower the vigour of the oak trees. However, our study cannot allow us to conclude that poor vigour or growth in a soil with low nutrient content could lead to an increased susceptibility to *C. fusipes*. In contrast, it is the better persistence of the pathogen on woody debris in the sandy acidic soils favourable to *C. fusipes* that clearly increases the disease risk. Because *C. fusipes* infection progresses at the root/soil interface by ectotrophic mycelial growth, soil conditions favouring pathogen survival could also have a strong positive influence on disease progression on single trees in the long term. Soil conditions probably play a major role in the infection process by favouring the pathogen itself.

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