Influence of Pea Cropping History on Disease Severity and Yield Depression

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ABSTRACT

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Field plot studies at five experimental sites were conducted during 1989–1991 to examine the influence of pea cropping history on disease severity and yield depression in peas (*Pisum sativum*). A modified greenhouse test for estimating the degree of infestation of fields proved to be a good predictor of disease severity. Several major pathogens were involved in the disease complex, and the total number of colonies isolated from the root systems and epicotyls appeared to be correlated with a disease severity index. In soils not previously cropped with legumes, two successive pea crops resulted in a slight yield decrease in the second-year crop. In naturally infested soils, the yield reduction was more pronounced. *Aphanomyces euteiches* was recovered from plants after two pea crops in 3 yr. This study suggests that cultivation with a conventional moldboard plow to a depth of about 20 cm cannot eliminate the increase in infestation of the root rot complex caused by a single pea crop. In soils with severe infestation of root rot pathogens, plant height seems to be closely correlated with the yield of dry peas. Aboveground symptoms, such as stunted plant growth, yellowing, and wilting, were only noticed for severely affected plants.

Additional keyword: Fusarium

Root diseases of pea (Pisum sativum L.) are an increasing problem for growers and pea processing companies in the Scandinavian countries. Several worldwide surveys have shown that pea root rot is caused by a complex of root pathogens (3,4,6,7,10,12,13,16,19,21,23, 25,31,33,35,36). In the United States, more than 25 fungi are listed as pathogens of pea roots (11). The lack of effective fungicides and an increasing awareness of environmental side effects of pesticides emphasize the need for nonchemical control measures, including soil prediction tests, cultural practices, and, especially, genetic resistance. The importance of individual pathogens varies in different geographic areas because of differences in environmental factors, cultural practices, and pea cultivars grown and is often based on a more or less subjective estimation. It is often difficult to relate discoloration of the root system and yield depression to a single pathogen in the root rot complex under field conditions because several pathogens can be involved and can interact with a complex of biotic and abiotic factors resulting in very indistinct symptoms. In most reports, the estimated prevalence of individual pathogens is based on the frequency of fields from which the respective fungi are isolated. However, Tu (33) managed to calculate a disease damage index for the four main root rot pathogens in Ontario, Canada, and rank their

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importance on the basis of the total amount of root rot, frequency of occurrence of each root rot fungus, and disease severity caused by each fungus.

Sherwood and Hagedorn (30) introduced a greenhouse technique for estimating the degree of infestation of fields by Aphanomyces euteiches Drechs. prior to planting. The technique has been modified and adapted in many pea-growing countries (4,7,17,21,23,27,31,33). Most reports show a linear regression between root disease severity and yield reduction. Basu and coworkers (1,2) investigated the relationship between yield loss and root rot severity in a field infested with Fusarium solani (Mart.) Sacc. f. sp. pisi (F.P. Jones) W.C. Snyder & H.N. Hans. They concluded that yield loss in growers' fields could be estimated by multiplying the percentage of moderately affected plants by a loss conversion factor of 0.23 (1) or the percentage of severely affected plants by 0.57 (2).

The objective of our study was to determine the influence of pea cropping history on disease severity and yield depression in dry pea in fields with different levels of natural infestation of root pathogens.

MATERIALS AND METHODS

Design of field plot experiments. Identical field experiments were conducted at five locations in Denmark: Tylstrup, Ronhave, Odder, Sejet, and Durup. The field histories of Durup, Ronhave, and Tylstrup did not include any legume crop for at least 30 yr. The fields at Odder and Sejet were naturally infested with root pathogens because of

previous pea crops. At the Odder site, peas were grown in 1980 and 1985. At the Sejet site, an unknown number of legume crops was grown in the field before the trials were established. The site at Tylstrup was a sandy soil, whereas the four other sites were sandy loam soils. Soil pH at Tylstrup, Ronhave, Odder, Sejet, and Durup was 6.6, 7.1, 7.6, 6.4, and 6.8, respectively, and the percentage of organic matter was 1.9, 2.0, 2.8, 2.4, and 2.5, respectively. The site at Tylstrup was irrigated when precipitation deficit was more than 30 mm. The field plot trials were initiated in 1989 and carried out over a period of 3 yr. Four replicated blocks were divided into two plots of 15 × 12 m, and one plot was planted with spring barley and the other with pea cv. Bodil. The plots were left over the winter, and in 1990 each plot was subdivided into two smaller plots $(7.5 \times 12 \text{ m})$, with one planted with spring barley and the other with pea. In 1991, pea was planted in all plots, resulting in four different combinations of pea: plot 1 = 1989barley, 1990 barley, 1991 pea; plot 2 = 1989 barley, 1990 pea, 1991 pea; plot 3 = 1989 pea, 1990 barley, 1991 pea; and plot 4 = 1989 pea, 1990 pea, 1991 pea. In the fall, each site was treated with a conventional moldboard plow to a depth of about 20 cm, and the seedbed was prepared and planted at the end of March. The peas were harvested at dry harvest stage (15) in August. The yield was adjusted to 15% water content. Thousand grain weight was measured only at the Durup, Odder, and Sejet sites. To avoid the influence of cross-contamination between adjacent plots, only 2.5 × 12 m of each plot was harvested and used for collection of soil and plant samples. Insecticide and fungicide treatments against airborne pests and diseases were applied if necessary.

Soil sampling and soil prediction test. The soil prediction test described by Biddle (4) was modified as follows: Before planting, 30 subsamples were randomly taken from the plow layer (20 cm) of the four plots and pooled into one soil sample (6 kg) for each combination of pea (four replicate plots). One soil sample in 1989, two soil samples in 1990, and four soil samples in 1991 were taken from each site. The prediction test was performed in the greenhouse, where the soil sample was thoroughly homogenized and transferred to three replicate disinfected plant pots (16 cm diameter). Pea seeds (cv. Bodil) were surface-disinfected

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in 1.5% (w/v) sodium hypochlorite for 8 min, washed three times in sterile water. and planted at a depth of 3 cm, 10 seeds per pot. The pots were watered each day to field capacity and kept at 18-21 C in the greenhouse supplemented with artificial light in a 16-hr day length (90 $\mu \text{E} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) for 5 wk. Disease severity of each individual plant was visually assessed as percentage of root and epicotyl length that was discolored, then scored into six classes, from 0 = no discoloration to 5 = complete discoloration. A disease severity index (DSI) was calculated separately for epicotyls and roots. The two score totals for all 30 plants were added and divided by two to give a predicted DSI (0-100).

Field disease severity index. To compare the predicted DSI in the greenhouse with the actual DSI in the field, five plants were dug at random from each plot at full flowering stage. The four plant samples on five plants from each combination of peas were pooled and scored the same way as described for the prediction test. An average DSI was calculated for all 20 plants.

Isolation of root pathogens. From the 20 pea plants collected from each combination of peas, five symptomatic plants were selected for isolation of root path-

ogens. The stem was removed at the secunder running tap water for at least 1 hr. The roots and epicotyls were surfacesystem with sodium hypochlorite, sections were transferred to the selective MBV medium (24) for isolation of A. euteiches.

RESULTS

Table 1 shows the total number of colonies of F. solani, F. oxysporum Schlechtend.: Fr., Phoma medicaginis Malbr. & Roum. in Roum. var. pinodella (Jones) Boerema, and Pythium spp. isolated from plants in plot 1 in 1991, along with the field DSI. A. euteiches could not be recovered on SNA and PDA media and was recovered on MBV medium from a smaller number of plants than showed the presence of the typical

ond node; the remaining root system was washed by gently rubbing with ordinary hand soap for 15-30 sec and then placed disinfected in 1.5% (w/v) sodium hypochlorite for 2 min and dried between pieces of sterile filter paper. From the interface between healthy and diseased tissue, small transverse sections were cut and transferred to plates with PDA and SNA media (20) amended with 25 ppm of tetracycline and 50 ppm of chloramphenicol. Before disinfection of the root

> only at Sejet. A significant regression between the predicted DSI, scored on test plants in the greenhouse, and the actual DSI, scored in the field, was recorded for four different combinations of peas at five different sites (Fig. 1, Table 2). The regression line intersects the y-axis at 35.9. which indicates the amount of discoloration due to abiotic factors.

oospores in the cortex by means of mi-

croscopic examination. Therefore, the

colony number of A. euteiches recovered

on MBV medium is omitted from Table

1 and the pathogen is recorded as present

(+) or absent (-). A. euteiches was pres-

ent in all five symptomatic plants in plot

1 at the Sejet site because of natural

infestation. The fungus was present in

plot 4 at four of the five experimental

sites, i.e., Ronhave, Durup, Tylstrup,

and Sejet. The fungus was present in all

plots at Sejet but absent in all plots at

Odder. The fungus was also present in

plot 2 at Tylstrup and plot 3 at Ronhave.

plants in the greenhouse and on plants

from field plots, along with yield of dry

peas, recorded in the field for the four

combinations of peas at the five exper-

imental sites. Thousand grain weight was

measured only at Durup, Odder, and

Sejet, and plant height was measured

Table 2 shows the DSI scores on test

On average, 1 yr with peas resulted in an increase in the predicted and field DSIs and in a significant yield reduction in the subsequent pea crop (plot 2) in two of the five trials. On average, two pea crops in 3 yr with a break crop of barley (plot 3) resulted in a slight increase in the disease severity index for the three sites not previously cropped with legumes. The corresponding lower yield at four of the five sites was significant (P < 0.05) at the sandy soil site at Tylstrup. On naturally infested soil at Odder, the predicted DSI increased from 31 to 98. The difference in the field DSI between plot 1 and plot 3 was not as large as estimated in the greenhouse test. At the Sejet site, the natural infestation was high and resulted in the maximum DSI for all plots in the prediction test.

Table 1. Number of colonies of Fusarium solani, F. oxysporum, Phoma medicaginis var. pinodella, and Pythium spp. and presence (+) or absence (-) of Aphanomyces euteiches oospores isolated from five symptomatic plants in plot 1 in 1991 at five experimental sites in Denmark

Site	F. solani	F. oxysporum				DSI ²	A. euteiches				
			P. m. var. pinodella	<i>Pythium</i> spp.	Total		Plot 1	Plot 2	Plot 3	Plot	
Ronhave	0	3	3	0	6	26	_		+	+	
Durup	0	4	2	0	6	40		_		÷	
Tylstrup	2	1	7	0	10	47	_	+	_	+	
Odder	2	4	11	6	23	76	_		_		
Sejet	7	11	17	Ö	35	86	+	+	+	+	

[&]quot;Isolated from roots and epicotyls on SNA and PDA media.

Table 2. Effect of four different combinations of peas in a 3-yr rotation on disease severity index predicted in the greenhouse and scored in the field and on seed yield of dry peas

	Ronhave				Durup			Tylstrup		Odder				Sejet					
	DSI*		_ Yield	DSI		Yield		DSI		Yield	DSI		Yield		DSI		Yield	Di 4	
Plot	Prd.	Field	(kg/ha)	Prd.	Field	(kg/ha)	Tgw ^x	Prd.	Field					Tgw	Prd.	Field	(kg/ha)	Tgw	Plant height ^y
1	18 a²		5,123 a			5,793 a	308 a	12 a	47	4.158 a	31 a	76	4 712 a	308 a	100	86	3 845 2	325	53 3 0
2	18 a	38	5,130 a	36 ab	50	5,539 ab	305 ab	37 bc	65	3.758 bc	73 b	86	4 009 a	203 a	100	90	3,133 bc	286	11.5 h
3	13 a	37	5,183 a	24 a	38	5,602 ab	300 ь	20 ab	56	3.923 b	98 b	84	4 542 a	306 a	100	100	3,678 ab	320	52.9 0
4	44 b	49	4,875 a	73 b	64	5,176 b	299 ь	48 c	76	3,363 c	96 b	93	2,785 a	279 a	100	100	2.630 c	277	32.6 a

Field plot design: plot 1 = 1989 barley, 1990 barley, 1991 pea; plot 2 = 1989 barley, 1990 pea, 1991 pea; plot 3 = 1989 pea, 1990 barley, 1991 pea; plot 4 = 1989 pea, 1990 pea, 1991 pea.

Determined by microscopic examination of the cortex.

Field plot design: plot $\hat{1} = 1989$ barley, 1990 barley, 1991 pea; plot 2 = 1989 barley, 1990 pea, 1991 pea; plot 3 = 1989 pea, 1990 barley, 1991 pea; plot 4 = 1989 pea, 1990 pea, 1991 pea.

² Disease severity index, where 0 = no discoloration and 5 = complete discoloration of root and epicotyl (see text).

^{*}Disease severity index, where 0 = no discoloration and 5 = complete discoloration of root and epicotyl (see text). Prd. = predicted in the greenhouse.

^xThousand grain weight, measured only at Durup, Odder, and Sejet.

y Measured only at Sejet, at full-flowering stage.

² Means followed by the same letter are not significantly different (P < 0.05).

In the field, only plot 1 showed a DSI below 100, which indicated a complete discoloration of epicotyl tissue and the entire root system (Table 1). The overall difference in the predicted and field DSIs observed at the Tylstrup site was due to a brownish discoloration of the outer cortex caused by water-soaking and secondary fungi.

Figure 2 shows the relationship between yield of dry peas and the field DSI for each of the five field trials (Fig. 2). Yield decreased dramatically in naturally infested fields at Odder and Sejet, where the field DSI in plot 1 was above 70. Yield at the three sites not previously cropped with peas (Durup, Ronhave, and Tylstrup) with a field DSI in plot 1 under 50 was less influenced by the 3-yr monoculture of peas. The relationship between disease severity seems to be linear for moderately affected plants and strongly curved for severely affected

Figure 3 shows the relationship between thousand grain weight and yield of dry peas for Durup, Odder, and Sejet. The linear regression was significant (P < 0.05) only for Odder and Sejet.

Plant height was recorded only at the Sejet site and was measured at the fullflowering stage when all plants were green. Later, at the pod-filling stage, all plants in plot 4 yellowed and wilted,

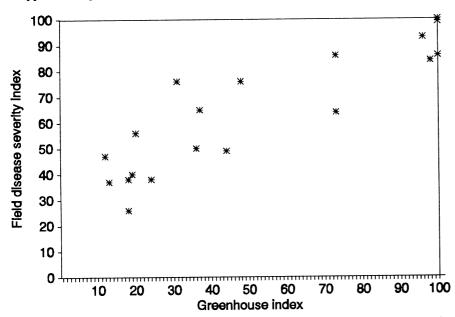


Fig. 1. The relationship between the field disease severity index and the predicted disease severity index for 20 soil and plant samples from five field trials with four combinations of peas in a 3-yr crop rotation.

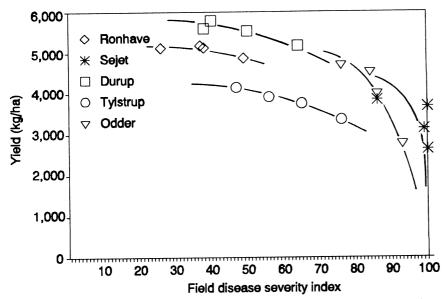


Fig. 2. The relationship between seed yield of dry peas and the field disease severity index in five trials (Ronhave, Sejet, Durup, Tylstrup, and Odder). In 1991, the field disease severity index was scored and the seed yield measured in all pea plots for four combinations of peas in a 3-yr crop rotation (1989 barley, 1990 barley, 1991 pea; 1989 barley, 1990 pea, 1991 pea; 1989 pea, 1990 barley, 1991 pea; 1989 pea, 1990 pea, 1991 pea).

whereas the plants in plots 1, 2, and 3 were still green, despite the completely discolored root system. There was a significant correlation between plant height and yield of dry peas and between plant height and thousand grain weight (Fig. 4). Plant height, like yield and thousand grain weight, was significantly influenced by disease severity (Table 2).

DISCUSSION

It is not common to grow successive crops of peas in the Scandinavian countries. However, in certain areas of Scandinavia, especially around processing companies, peas are grown quite intensively in a rotation with only a 4-yr break between pea crops. This results in a strong need for reliable methods of predicting disease potentials for specific fields. In our effort to implement a prediction system under Danish conditions, we have tried to illuminate some of the aspects of root diseases of peas and their effects on yield by growing different combinations of peas in a 3-yr rotation. Studies of epidemiology of soilborne pathogens have shown a clear relationship between inoculum level and rate of disease increase (8,25,26). In peas, the rate of disease increase due to A. euteiches has been shown to be similar for all inoculum levels. A low inoculum level only influences the time required to reach 50% disease incidence (25). However, our study indicates that the time from planting to 50% disease incidence might be crucial for the extent of yield loss. This was apparent especially at the Sejet site. Unfortunately, the disease development in the plots at Sejet was not followed from planting to flowering, but at the time of flowering, a small difference in DSI in the field resulted in a considerable difference in yield. The rate, delaying, and importance of the disease increment by different inoculum levels might be different when looking at a complex of root pathogens rather than a single member of the complex. This aspect needs further investigation.

The modified greenhouse test for estimating the degree of infestation of fields prior to planting appears to be a reliable technique (Fig. 1). Because of different environmental conditions in the field, accuracy of the soil test can vary between years. However, variation for severely infested fields is less than that of moderately infested fields, and a low DSI in the greenhouse never leads to a high DSI in the field (unpublished). Classification of the mild root rot symptoms in Figure 1 must, however, be adjusted for discoloration caused by abiotic factors.

Several pathogens are involved in disease development. The total number of colonies of four of the major pathogens isolated from diseased pea tissue was correlated with the DSI recorded in the field (Table 1). This is in agreement with

Tu (33), who found that the overall disease severity and yield loss were the sum contributed by each component fungus of the pea root rot complex in Ontario, Canada. Whalley et al (35) found a significant correlation between the predicted DSI and the individual and combined populations of P. m. pinodella and F. solani by means of a dilution plate method. Many reports focus on a single pathogen as responsible for the root rot in a particular pea-growing area. From the results of this study it seems likely that a single or a few pathogen species are involved at low levels of disease severity, whereas several taxonomic groups of pathogens seem to be involved at higher levels.

This study suggests that a single crop of peas increases infestation of pea root rot pathogens (Table 2). The increase in disease severity measured at full bloom is very much influenced by the initial level of infestation due to pea cropping history. At the experimental sites at Durup and Tylstrup, which had not previously been cropped with legumes, two successive pea crops resulted in a slight yield decrease in the second-year crop. In naturally infested fields at Odder, the yield reduction was more pronounced. Three successive years in peas led to a yield reduction of 5-41% at all five experimental sites. Salt and Delany (29) found a 24% yield reduction in the straw yield in the fourth year of monoculture of peas at one experimental site.

A. euteiches, the most destructive root pathogen of peas in Scandinavia, was recorded in the third successive pea crop at four of the five experimental sites and after two pea crops in 3 yr at two experimental sites. We have several examples from a 3-yr disease survey in Denmark (unpublished) of the presence of A. euteiches in fields cropped with peas only twice in 10 yr. The same survey illustrated that a field infested with A. euteiches led to complete crop loss in the third pea crop in 17 yr. The greenhouse test was a reliable method to predict the level of infestation of root pathogens, including this very important pathogen. Even though the greenhouse test is expensive and laborious, the cost is minimal compared to that of a severe crop

Most reports describe the time that is insufficient to reduce highly infested fields to a safe low level. Zogg (37) estimated 5-6 yr; Sundheim and Wiggen (31) and Temp and Hagedorn (32), 6-8 yr; Pfender and Hagedorn (25), 9 yr; Jones and Linford (14), Davis and Shehata (9), and Biddle (5), 10 yr; and Oloffson (22), 15 yr as insufficient to reduce soil inoculum to a safe level. Our experience from this study and a 3-yr disease survey in Denmark has shown that it is impossible in practice to predict the number of pea-free years that are necessary to avoid root rot. Disease severity and seed

yield of dry peas are very much influenced by the level of soil infestation and composition of root rot pathogens in each individual field resulting from the pea cropping history.

Rush and Kraft (28) found no infected pea roots in a greenhouse test when inoculum of F. s. pisi was placed in the lower 10 cm of containers 30 cm deep. They suggested that in the absence of other stress factors, inoculum of F. s. pisi located deep in the soil had no detrimental effect on pea growth and development up to the time of flowering, when the upper 20 cm of the root system is free of infection. A later study (18) showed the importance of the interaction of a tillage pan and the severity of F. s. pisi. When the tillage pan restricted pea roots to the top layer of soil, Fusarium root rot could be severe regardless of

amount of inoculum. Our study suggests that plowing to a depth of 20 cm with a conventional moldboard plow cannot reduce the infestation after a single pea crop when the overall root rot complex is considered, and not just F. s. pisi as a single member of the complex. Furthermore, keeping the upper 20 cm free of infection will be difficult because the inoculum will be mixed up in the plow layer owing to the usual soil preparation during a normal 4-5 yr break. This strategy is therefore not applicable in Scandinavia. In areas where peas can be grown in succession, however, field experiments are needed to determine if this strategy, which must include subsoiling. can be successful at different levels of natural infestations under commercial conditions.

Thousand grain weight was correlated

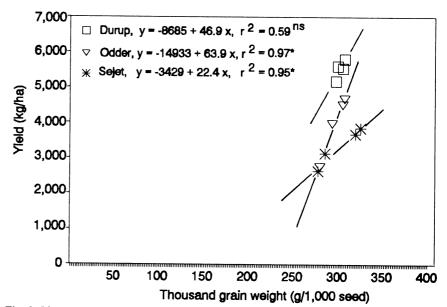


Fig. 3. Linear regression between thousand grain weight and yield of dry peas at three sites (Durup, Odder, and Sejet) in four combinations of peas in a 3-yr rotation.

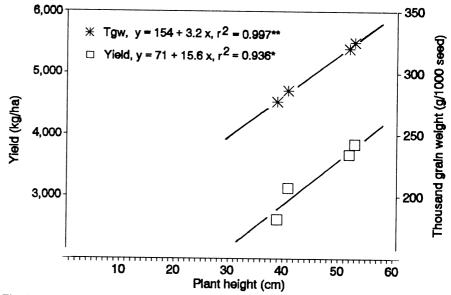


Fig. 4. The relationship between plant height and yield of dry peas and between plant height and thousand grain weight at the Sejet site.

to the field DSI (Fig. 3). The decrease in thousand grain weight can explain only some of the yield decrease. Because damping-off, caused by *Pythium* spp. at Sejet and Durup, was insignificant, the number of plants might not have been reduced significantly. The increased disease severity may therefore have reduced the number of seeds in the pods.

Plant height seems to be better correlated with yield than with thousand grain weight (Fig. 4). Aboveground symptoms, such as chlorosis and wilting, were seen only in plot 4 at Odder and Sejet. This is in agreement with Basu (1), who found that only severely affected plants showed symptoms. The development of aboveground symptoms is influenced by soil moisture. If the soil structure is loose and the plants are supplied with adequate water and nutrition, aboveground symptoms sometimes do not appear even in severely infected plants.

The pea germ plasm collections contain lines with moderate to high resistance against most pathogens in the root rot complex. In most surveys of pea root diseases, F. oxysporum was isolated in high frequency as a component of the root rot disease complex (12,29,35). Tu (34) tested a large number of commercial cultivars for race-specific resistance to Fusarium wilt and race-nonspecific resistance to Fusarium root rot in an infested field and found different degrees of susceptibility. Durable disease resistance in legumes has not received much attention. Cultivars with a high level of durable resistance are very valuable for the worldwide market and for pea growers with a limited knowledge about the complex character of soilborne pathogens in the field. Greater efforts are needed for screening peas for both racespecific and race-nonspecific resistance against pathogens involved in the root rot complex in peas.

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