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A comparison of two pathosystems: downy mildew on Spinacia oleracea and on Chenopodium album

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Abstract

The development of downy mildew *(Peronospora farinosa)* on a cultivated spinach crop *(Spinacia oleracea)* was compared to that on lambsquarters *(Chenopodium album)*, a naturally occurring weed. Experiments were carried out on small spinach plots, in which epidemics could develop from point sources (PS) or area sources (AS).

Two lambsquarter populations were studied, in which downy mildew epidemics arose from oospore infections. The downy mildew epidemics, on spinach and lambtquarters, were related to the environmental factors: temperature, vapour pressure deficit (VPD) and rainfall. It is concluded that the two different formae speciales of *P. farinosa,* one on spinach and one on lambsquarters, react in the same way to weather changes. The epidemiological patterns were almost similar.

Additional keywords: Peronospora farinosa, epidemic.

Introduction

Lambsquarters (= fat-hen; *Chenopodium album* L.) is a common species of the Dutch flora. The general occurrence and nutritional value of this weed made it a vegetable in the Dutch cuisine of the past (Zeven and Zhukovsky, 1975). Later it was replaced by spinach *(Spinacia oleracea* L.), which had a better taste and higher yield. Nowadays *C. album*, considered as a weed, is frequently found in many crops.

Beet, lambsquarters and spinach are attacked by the same species of downy mildew *(Peronospora farinosa* (Fr.) Fr.). Several authors tried to cross-inoculate the downy mildews of these plant species without success. This led to the recognition of three formae speciales, each with a specific and narrow host range (Byford, 1967): f. sp. *betae* on *Beta* sp., f. sp. *spinaciae* on *S. oleracea* and f. sp. *chenopodii* on *C. album.*

The purpose of this study is a comparison of epidemics of the pathogens *P. farinosa* f. sp. *spinaciae* and *P. farinosa* f. sp. *chenopodii*, the former on a cultivated crop (spinach), in which the fungus was artificially introduced, and the latter on a naturally occurring species (lambsquarters), in which the disease appeared spontaneously. Epidemics can be compared by means of the factors by which they are characterized:

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the initial inoculum and the infection rate. The infection rate is governed by the duration of the latency period, the duration of the infectious period, the effectiveness of the inoculum to produce new infections, and the rate of spore production (Zadoks and Schein, 1979). These parameters are more or less influenced by climatic conditions, crop density and structure, as well al plant and pathogen genetics. The infection rate itself, therefore, is not constant during the growing season.

As the experiments with spinach and lambtquarters were set up for other purposes than comparison, the epidemiological parameters did not get the attention needed for a complete comparison. Where possible they will be used in the description of and discussion on the epidemics.

As far as known to the authors the comparison between 'one' pathogen and two closely related plant species, with different backgrounds in cultural practice, has not yet been made. It can provide valuable information about the characteristics of downy mildew epidemics in crops in relation to weather and crop development, and it can enlarge our understanding of complex pathosystems (Kranz, 1974, 1978, 1980).

Materials and methods

Spinach

Field. The epidemics of downy mildew were followed in 25 miniplots of 1.2 × 1.2 m. These plots were sown with spinach cv. Noorman, susceptible to pathotypes 1, 2 and 3. Sowing was done by hand in rows of 1.2 m length and a distance of 0.15 m between rows. Each plot was separated from its neighbour plots by a buffer zone of 4 m, in which the resistant spinach cv. Wolter was sown.

Plant growth. The growth of the spinach plants was followed by measuring the leaf area in cm^2 of 25 plants randomly chosen from a plot not in use for the inoculation experiments.

Inoculation. The plots were inoculated with a suspension of conidia of *P. farinosa* f. sp. *spinaciae* pathotype 3 by means of a DeVilbiss atomizer. Inoculum density was 8.10^4 conidia ml^{-1}.

Two types of inoculation could be distinguished: area source inoculations (AS) and point source inoculations (PS). In case of AS inoculations each plot was inoculated with 75 ml inoculum; in case of PS inoculations, only three plants in the center of the plot were inoculated with a proportionally smaller amount of inoculum. Inoculations were scheduled at sunset to ensure relatively low temperatures and high air humidities ($>80\%$) for infection. The AS as well as the PS inoculations were carried out at different development stages of the plant. One third of the plots was inoculated when plants had their cotyledons completely developed (ASc and PSc), one third when the plants had their first pair of true leaves (AS1 and PS1), and one third when they showed their third pair of true leaves (AS3 and PS3).

Disease assessment. Observations took place in triplicate for each treatment. They started directly after the expiration of the first latency period. Disease severity was defined and estimated according to Clive James (1974) as the diseased leaf area expressed as a fraction of the total leaf area. Total leaf areas were measured by comparison of the leaves to standard diagrams of leaf area.

Environmental data. Temperature, relative air humidity (RH) and rainfall were recorded by thermocouples at plant level, a thermohygrograph and a standard rain gauge. RH data were converted to vapour pressure deficit (VPD) according to Stevens' (1916) formula:

$VPD = 1 - \frac{RH}{100}).E_T$, in which VPD is the vapour pressure deficit in mm Hg, RH is the relative humidity in %, and E_T is the vapour pressure at saturation at a given T in mm Hg. VPD is a more meaningful expression in case of changing temperatures.

Common lambsquarters

Field. The epidemics of downy mildew in lambsquarters were followed in two naturally occurring populations (A and B) in and at the border of a spinach crop, situated at about 4 km from the spinach experiment. The populations covered areas of about 1.5 × 2.5 m.

Plant growth. The growth of the lambsquarters was followed by estimating the mean plant height in cm.

Inoculation. No artificial inoculation was carried out. The epidemics appeared spontaneously. Plants of 3 to 6 cm length were present, at a density of 100 plants per m^2, when sporulating showed up first on a primary leaf of a single plant, belonging to the category of smallest plants.

Disease assessment. For disease assessment the area covered by the population under observation was divided into squares of 0.25 × 0.25 m. In these squares disease intensity was estimated as disease incidence according to the definition of Clive James (1974). The number of diseased plants was counted and expressed as fraction of the total number of plants.

Environmental data. No weather data were recorded in the observed plots. For the interpretation of the results the data from the spinach field at a distance of 4 km were used.

Results

Weather. Fig. 1a shows the course of three main climatic factors during the period between 4 May and 7 July 1982: day 08.00 am till 10.00 pm and night 10.00 pm till 08.00 am, air humidity as VPD, day and night temperature and rainfall. The periods of interest (I through VIII) are described shortly in Table 1.

Plant growth. Leaf area development (spinach) as well as plant growth (lambsquarters) are shown in Fig. 2. The emergence of lambsquarters was regular in both populations A and B. These two populations of young plants were dense and grew in areas of 0.5 to 1.0 m by 1.0 to 2.0 m. Outside these areas the populations were less dense. Both,

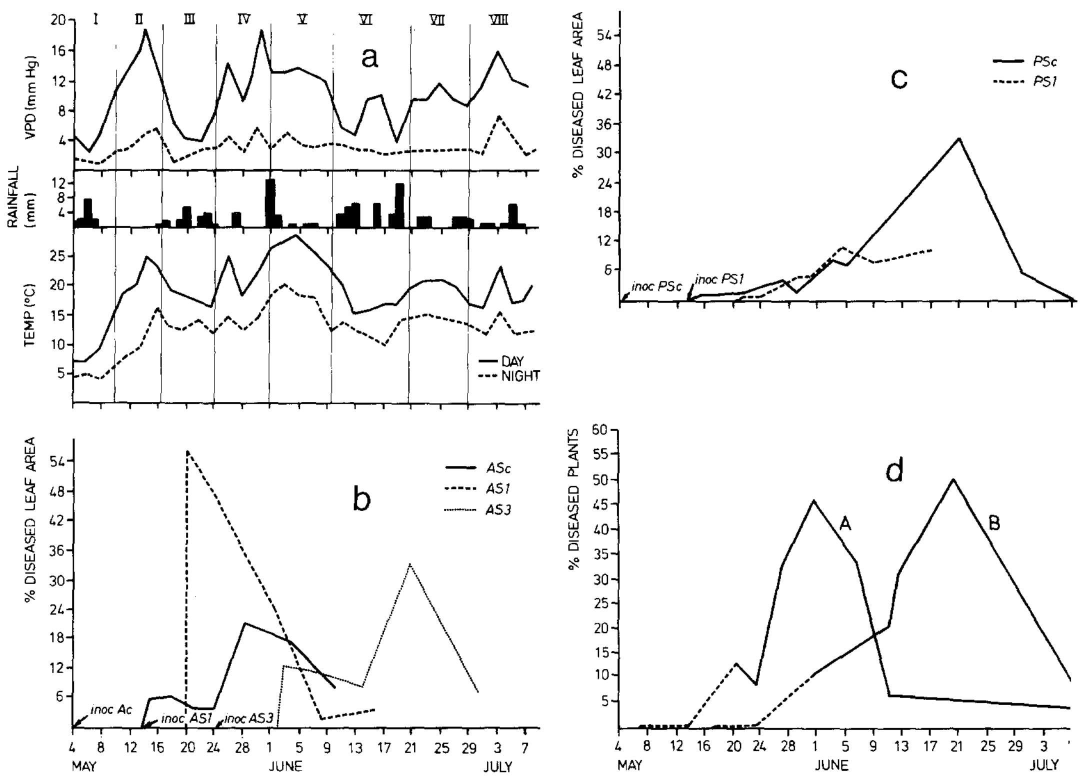

Fig. 1a. The environmental factors vapour pressure deficit (VPD), mean temperature, both during day and night, and rainfall. The time span from 4 May till 7 July 1982, over which weather was recorded, is divided into periods (I to VIII) corresponding with those of Table 1.

Fig. 1b and 1c. Development of spinach downy mildew in % diseased leaf area. Inoculation in different plant stages: cotyledons (c), first leaf pair (1) and third leaf pair (3). Inoculations were either area source (AS) or point source inoculations (PS).

Fig. 1d. Development of downy mildew on two populations of lambsquarters (A and B) expressed in % of total number of plants. The period of observation was 4 May till 7 July 1982.

lambsquarters of population A and spinach remained in the seedling stage until half May. The plants of both species developed vegetatively until the beginning of June by producing new leaves. Hereafter stem elongation began. Flowering started by half June followed by an accelerated decrease in total leaf area for spinach. Cotyledons dropped or decayed. Lambsquarters dropped the leaves attacked by downy mildew. Seed set began at the end of June in both spinach and lambsquarters.

The main differences between the two species are the more pronounced stem elongation and the greater diversity in ultimate plant height in lambsquarters as compared to spinach.

Lambsquarters of population B showed almost the same growth and development pattern as population A, but with a delay of 2 to 3 weeks.

Table 1. Short description of weather types during eight consecutive periods of observation (D = day; N = night).

Period	Weather description
Period I 4.5-10.5	cold and wet D and N temperatures below 15 °C (= mean N temperature over the whole observation period); VPD values low; frequent rainshowers
Period II 11.5-17.5	high D temperatures and increasing N temperatures; VPD values relatively high (D and N); little rainfall on 16 May
Period III 18.5-24.5	moderate weather type; D and N temperatures constant between 12 and 18 °C; VPD values low; frequent rainshowers
Period IV 25.5-1.6	relatively warm and dry but alternating; VPD values alternating high and low; short and heavy rainshowers on 27 May and 1 June
Period V 2.6-10.6	hot period with some 'summerly' days (> 25 °C); VPD values constant and relatively high during daytime; little rainfall
Period VI 11.6-21.6	weather type comparable with third period; VPD values low, except in the middle of the period (15 and 16 June); frequent rainshowers
Period VII 22.6-29.6	steady weather type; D and N temperature moderately high; VPD values stable; isolated rainshowers
Period VIII 30.6-7.7	relatively high temperatures (falling after 3 July); VPD values high (falling after 3 July); little rainfall (except 5 July)

Description of epidemics

Disease progress of downy mildew epidemics on spinach with area source inoculation (ASc, AS1 and AS3), on spinach with point source inoculation (PSc and PS1) and on lambsquarters (populations A and B) is described separately (Figs 1b, c, d). The PS3 epidemic could not be used for comparison as it started before artificial inoculation could take place. This was probably caused by the introduction of conidia from other plots.

ASc – AS1 – AS3 (S. Oleracea). The first sporulation wave of ASc became visible after a latency period of 11 days, a relatively long period because of the low

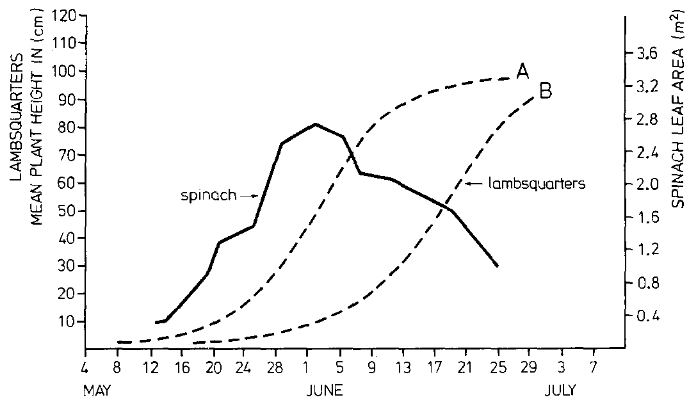

Fig. 2. Development of cultivated spinach and of two spontaneous populations of lambsquarters (A and B), both over the period 4 May till 7 July 1982.

temperatures during the first period. There was no further disease progress till 24 May, when a second sporulation wave appeared with a peak on 28 May. There was an increase of sporulating leaf area till 4 June, but as total leaf area increased too between 31 May and 4 June, the increase in sporulating area is translated into a decrease of disease severity on the graph.

The first sporulation of AS1 was observed on 21 May after a latency period of 7 days, which is normal under near-optimal weather conditions. The disease severity was extremely high (> 50%), but dropped drastically to a few percent on 8 June, followed by a slight revival, as for a second wave. The amount of disease was constant between the first observation and 3 June.

The first latency period of epidemic AS3 was 9 days, ending 3 June. Both the disease severity and the amount of disease remained constant till 14 June. Between 3 and 14 June the first wave showed up. Thereafter, both disease amount and severity increased to form a second wave with a maximum at 21 June. Then, the epidemic declined and nearly disappeared at 7 July.

PSc – PS1 (S. oleracea). The epidemics initiated by point sources (PSc and PS1) had the same latency period as ASc and AS1, viz. 11 and 7 days. The disease progress was slow in the beginning. It was characterized by two epidemic waves with their maxima at 27 May – 3 June and 28 May – 4 June, respectively. For PSc a third wave with a maximum at 21 June looked more impressive than the first two waves, but its height is due in part to a decrease of leaf area. Remarkable is the date of the maximum, which is the same as the date for the top of the second AS3 wave.

Populations A and B (C. album). The disease on common lambsquarters' population A was detected on 21 May, when 14% of the plants were attacked by downy mildew. It was about the peak of the first wave. The exact starting date of the epidemic was difficult to indicate, for the epidemics on lambsquarters emerge from an oospore infection in a young stage of the population. The plant which served as initial inoculum source could be identified easily, for it lagged behind in development (Fig. 3). The

Fig. 3. Plant within a lambsquarters population, infected by oospores, serving as a point source in the development of *Peronospora farinosa* f. sp. *chenopodii.*

epidemic showed a second wave with a maximum around 1 June. Hereafter, practically no new plants were infected. Instead, small roundish local lesions appeared on the upper leaves of the plants. The epidemic decreased rapidly to a low level after 12 June, since most plants dropped their infected leaves. The epidemic in population B showed the same course. It reached a maximum on 21 June after which date the roundish lesions appeared as in population A. Remarkable again is the date of 21 June.

Discussion

Initial inoculum. The origin of the primary infection in common lambsquarters differed from the one in spinach. Spinach was artificially inoculated with conidia, both in the AS and the PS plots. In Lambsquarters the primary infection had a natural origin. In populations A and B as well as in most of the locations observed for other purposes the source plant(s), from where the epidemic began, could be indicated. In the centre of a focus one or more seedlings were found with chlorotic, deformed, and heavily sporulating leaves. The obvious explanation for these symptoms is an infection by oospores. This has been recorded earlier for many pathosystems of which members of the Peronosporaceae family are a part (Spencer, 1981). Oospores of *P. farinosa* were claimed to be the most important source of primary infection in sugarbeet in the Netherlands (Van der Spek, 1964), and in spinach in the USA (Wright and Yerkes, 1950). The possible importance of oospores as a primary source of infection in spinach is accentuated by data showing that oospores can easily be produced in spinach cotyledons in laboratory and field experiments (Frinking et al., 1985). These oospores easily germinate and infect young spinach seedlings (Frinking, unpublished).

Oospore infections usually appear as point source. So, it should be possible to compare the epidemics on lambsquarters to those emerged from the PS inoculations of spinach. The quantity of initial inoculum, however, was not quite the same: in the A and B epidemics (lambsquarters) one oospore led to one sporulating plant, and in the PS epidemics (spinach) many conidia led to three sporulating spinach plants.

Latency period. This parameter is strongly related to weather conditions, especially to temperature. The latency period for spinach after infection by conidia lasts 7 days when conditions are optimal (AS1) (Frinking, unpublished). After the inoculations of the ASc/PSc and AS3 plots the duration of the latency period was 11 and 9 days, respectively. The relatively long duration for the ASc/PSc plots was due to low day and night temperatures. The first latency period for the epidemics of lambsquarters is estimated to be $\geqslant 14$ days. The length of this period is conditioned by the length of the germination time of oospores, which in laboratory experiments, was recorded as lasting 6 days or more (Frinking, unpublished). The time between initiation of oospore germination and sporulation is comparable to that of conidia infection: 8 to 9 days.

Patterns of epidemiological development. Optimal germination of spores of *P. farinosa* on spinach occurs between 5 and 15 °C and at about 25 °C (Frinking et al., 1981). The maximum temperature is 30 °C. For sporulation the VPD must be $\leqslant 0.6$ mm Hg (Kröber, pers. comm.). With these data in mind we will discuss the disease development during each of the weather periods described in Table 1.

The first weather period, with night-frosts was too cold for a successful infection in the ASc plots. Despite the cold weather the latency period of the fungus on the *C. album* seedlings in population A probably ended in this first period. After termination of a latency period of 11 days in the ASc and PSc plots, that was at the end of the second weather period, VPD values were high both during day and night. This prohibited an abundant sporulation and consequently a development on a high disease level. The temperatures however, were high enough for a good infection in the AS1 and PS1 plots, as indicated by the short latency period of 7 days.

The third weather period was optimal for infection and sporulation. The AS1 and PS1 plots began to sporulate and the first new infections on *C. album* in population A were detected. The *C. album* seedlings, functioning as initial inoculum in population B, probably began to sporulate in this period.

The fourth weather period was also suitable for infection and sporulation. New waves were observed in all epidemics. The ASc and A epidemics reached their maximum. Both the PS and B epidemics steadily increased.

The fifth period was too hot and dry for further progress in many epidemics. The infection levels in all AS plots and A decreased drastically.

In general, the weather conditions in the sixth period were suitable for a revival, which was observed in nearly all epidemics including AS1, in spite of the decrease in total leaf area for spinach. The revival climaxed on 21 June, on which date AS3, PSc and B reached their maximum. After this date all epidemics declined.

The *P. farinosa* formae speciales on spinach and on lambsquarters reacted similarly to changes in weather conditions. Changes in the epidemiological patterns were almost simultaneous.

Epidemiologically, the start of the epidemics was similar for the PS plots and the lambsquarters population. Both began with a small number of sporulating lesions. The shapes of the curves also show much similarity. In both cases we found a gradual build-up of the disease to a maximum, after which the epidemic decreased drastically. The infection rate, however, seemed to differ. The first sporulation in A started around 18 May and in PSc one week later. Both spinach and lambsquarters were still in the seedling stage. Epidemic A reached its maximum at 1 June and had a faster build-up than PSc which reached its maximum at 21 June.

The epidemics in PSc and B showed more similarity. The first sporulation of the infected seedlings in B occurred at the same time as the first sporulation of the artificially inoculated seedlings in PSc. Both epidemics reached their maximum on 21 June, although lambsquarters were less developed than spinach.

The AS plots showed different shapes of the epidemic curves. Disease progressed by leaps. The difference in disease progress between PS1 and AS3 after 3 June is interesting. In PS1 the disease increased gradually with a small decline from 3 to 5 June. In AS3 the increase was followed by a more important and more prolonged decline. Both epidemics reached a maximum of comparable height on the same date.

Epidemic decline. The end of the epidemics in spinach occurred at the end of the season, when total leaf area was decreasing and plants started to set seed. For *C. album,* however, the decline of the epidemic began before stem elongation and just before flowering.

In A, 49% of the plants were attacked by downy mildew when the epidemic reached its peak on 1 June. On 6 June practically no new sporulating lesions appeared, but on 63% of the plants, especially on the upper leaves, small roundish non-sporulating lesions could be observed. The same phenomenon occurred in B after 21 June. These symptoms suggest that the plants become hypersensitive to downy mildew when they reach a certain growth stage. The decreasing susceptibility with plant age was recorded for several downy mildews (Populer, 1978). In beet, each pair of leaves looses its susceptibility to *P. farinosa* f. sp. *betae* with growing age (Leach, 1931; Weltzien and Mey, pers. comm.). The leaves near the vegetation point remain susceptible (Weltzien and Mey, pers. comm.).

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Samenvatting

Een vergelijking van twee pathosystemen: valse meeldauw op Spinacia oleracea en op Chenopodium album

De ontwikkeling van valse meeldauw *(Peronospora farinosa)* op het cultuurgewas spinazie *(Spinacia oleracea)* werd vergeleken met die op het zich natuurlijk ontwikkelende onkruid melganzevoet *(Chenopodium album).* Daarvoor werden experimenten uitgevoerd op spinaziepercee ltjes, waarop in enkele gevallen punt-

broninfecties (PS), en in andere gevallen oppervlaktebrondinfecties (AS) waren aangebracht. Er werd gevarieerd in inoculatietijdstip.
Bij melganzevoet werden twee populaties bestudeerd, waarin de ziekte via oöspore-infectie op natuurlijke wijze was ontstaan.

Het geheel werd in verband gebracht met de omgevingsfactoren: temperatuur, dampdruk-deficiet (VPD) en regenval. Geconcludeerd kan worden, dat de *P. farinosa* formae speciales zowel op spinazie als op melganzevoet op gelijke wijze reageerden op veranderingen in het weer. Het verloop van de epidemieeën was sterk vergelijkbaar.

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