

EVALUATION OF FUNGICIDE APPLICATION
AT LATE BLOOM FOR THE CONTROL OF SCLEROTINIA
STEM ROT IN RAPESEED (CANOLA)

A Thesis

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Abstract

According to current recommendations, foliar fungicides for the control of sclerotinia stem rot in rapeseed should be applied at early or full bloom but not at late bloom. Field experiments were conducted in several areas of Saskatchewan in 1987 and 1988 to determine if spraying with benomyl at late bloom could adequately control the disease in at least some situations. The experiments were designed to determine if the growth stage at which inoculum of S. sclerotiorum first becomes widespread in the crop affects the efficacy of late bloom application. Rate of application and nozzle type were varied in a few tests.

Disease levels were high enough to warrant collection and analysis of disease incidence data at 4 of 5 locations in 1987 and 5 of 10 locations in 1988. Late bloom application of benomyl at the recommended rate (0.6 kg a.i./ha) significantly reduced disease incidence compared to the check in all tests except one. In four tests, disease reductions of $\geq 80\%$ were achieved. Generally, the highest levels of control were achieved when high levels of inoculum were not present in the crop until at least full bloom. In these situations benomyl was applied at approximately the same time that most infections were initiated. Application at 0.3 kg a.i./ha caused significant reductions in disease compared to the check at 3 of 4 locations, indicating that it may be

possible to control stem rot adequately by spraying at less than the recommended rate. Significant control was also achieved at 2 locations by spraying at 0.15 kg a.i./ha. There was limited evidence that small spray droplets generated by 11001 nozzles may improve control with benomyl.

The effect of time of infection (ie. plant growth stage) on yield loss was studied in growth chamber and field experiments. It was found that plants infected at late bloom suffer a significantly lower yield reduction than those infected earlier in the flowering period. In a field experiment, the yield reduction per plant was only 8% when high inoculum levels were not present until late bloom. More research is required in this area before fungicide application at late bloom can be considered an economically feasible option for rapeseed growers.

Finally, the disease incidence/disease severity (I-S) relationship for sclerotinia stem rot was studied using regression analysis. The slopes of regression equations were similar between years and, with the exception of one field, were similar among fields. This suggests that factors such as rainfall, stand density and inoculum levels have little effect on the I-S relationship. Fungicide application also did not appear to have an effect. The equation disease severity = $0 + 0.692 \times \text{disease incidence}$ described the overall relationship ($R^2=95.5\%$). This could be used to predict severity from incidence data, at least within Brassica napus cv. Westar, thereby simplifying disease assessment.

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1. Introduction

Spring rapeseed (canola) is an extremely important crop in western Canada. Both Brassica napus (Argentine-type rapeseed) and B. campestris (Polish-type rapeseed) are widely grown. One of the most important diseases affecting rapeseed is stem rot caused by Sclerotinia sclerotiorum (Lib.) de Bary [syn. Whetzelinia sclerotiorum (Lib.) Korf and Dumont]. The disease can cause substantial yield and quality losses when environmental conditions favor an epidemic. In 1982 sclerotinia stem rot of rapeseed in western Canada caused losses estimated to be in excess of \$15 million (Martens et al., 1984).

S. sclerotiorum has a worldwide distribution and an extremely wide host range which includes over 360 species in 64 plant families (Purdy, 1979; Willetts and Wong, 1980). The nonspecific nature of the pathogen has made breeding for resistance very difficult (Krüger, 1980; Verma, 1983). No Canadian rapeseed cultivars are resistant to the disease.

Cultural control methods include crop rotation, use of clean seed, deep plowing, weed control and burning of infected stubble. These methods decrease the number of viable sclerotia in the soil, thereby reducing the infectious potential of S. sclerotiorum. However, long distance dispersal of Sclerotinia ascospores and the longevity of sclerotia in soil limit the effectiveness of cultural control

methods (Morrall and Dueck, 1982; Williams and Stelfox, 1979, 1980).

Foliar application of fungicides is currently considered the best method of controlling sclerotinia stem rot (Davies, 1986; Morrall et al., 1985; Thomson et al., 1984). However, spraying is costly and must be carefully evaluated in terms of potential increases in yield. Growers must rely on disease forecasting methods to identify crops where economic returns from chemical control are probable (Thomas, 1984; Turkington, 1988). A quantitative forecasting system based on ascospore infestation of petals at early bloom is currently being studied and promising results have been obtained (Gugel and Morrall, 1986; Kaminski, 1987; Turkington, 1988). This system is thought to be superior to traditional forecasting methods based on the presence of apothecia, crop stand and disease history because it accounts for ascospores that move into rapeseed crops from extrinsic sources (Gugel and Morrall, 1986; Turkington, 1988).

In 1986 many areas of Saskatchewan (eg. Melfort area) were very dry in June and very few sclerotia germinated to form apothecia (Turkington, 1988). In almost all rapeseed crops sampled in these 'dry areas' the percentage petal infestation at early bloom was extremely low. The crops were considered to be at low disease risk and growers did not apply fungicides. However, improved moisture conditions in July

avored late inoculum production and infection. Consequently disease levels were moderate or high in a few fields.

Research has shown that aerial fungicide application is effective at early bloom and ground application is effective at early or full bloom (Dueck et al., 1983; Morrall et al., 1984b). Effective control with late bloom application has never been shown. The fungicides used against Sclerotinia have little therapeutic effect (Hunter et al., 1978; Steadman, 1979). Thus, late bloom application would not be expected to be effective if most infections were initiated earlier in the bloom period. In years such as 1986 fungicide application at late bloom might be effective because infections would be initiated at approximately the same time that the chemical was applied.

The primary objective of the present study was to evaluate the efficacy of fungicide application at late bloom in field experiments. Experiments were designed to determine if the crop growth stage at which significant amounts of inoculum first appear in the crop is a factor affecting the efficacy of late bloom application. The rate of chemical applied was varied in a few tests to see if significant control could be achieved by spraying at less than the recommended rate.

Another objective of this study was to investigate the effect of time of infection (ie. plant growth stage) on yield loss. If infections initiated late in the bloom period have

little effect on yield then late bloom fungicide application probably would not be economical. Field and growth chamber experiments were performed to meet this objective.

Finally, the disease incidence / disease severity (I-S) relationship for sclerotinia stem rot was studied using regression analysis. In several fungicide tests both disease incidence and disease severity values were calculated for each plot. The disease incidence value was simply the percentage of infected plants. The disease severity value was an index calculated after rating individual plants for amount of disease. A strong and consistent I-S relationship would enable researchers to omit disease severity ratings without a loss of important information.

2. Literature Review

2.1 Disease cycle of Sclerotinia sclerotiorum

2.1.1 General review

Stem rot is a soil-borne disease (Martens et al., 1984). S. sclerotiorum overwinters mainly as sclerotia in, or on, the soil and stubble, or as sclerotia with the seed (Thomas, 1984). Sclerotia can remain viable in soil for several years (Adams and Ayers, 1979; Cook et al., 1975). Under favorable conditions some of these sclerotia germinate to produce either hyphae (myceliogenic germination) or small mushroom-like apothecia which release air-borne ascospores (carpogenic germination) (Abawi and Grogan, 1979; Willetts and Wong, 1980). Most rapeseed infections result from ascospores; myceliogenic germination is relatively unimportant in the disease cycle (Kolte, 1985; Morrall and Dueck, 1982).

Carpogenic germination of sclerotia requires prolonged moist soil conditions and stable temperatures (Grogan and Abawi, 1975; Morrall, 1977; Willetts and Wong, 1980). Normally these conditions occur only if a dense plant canopy is present to completely shade the soil surface (Akai, 1981; Boland and Hall, 1987; Schwartz and Steadman, 1978; Morrall and Dueck, 1982). Morrall and Dueck (1982) observed that

apothecia never developed in a rapeseed crop before it was in the bud stage.

Apothecia forcibly discharge large numbers of ascospores which can travel considerable distances (150m or more) in air currents (Abawi and Grogan, 1979; Schwartz and Steadman, 1978; Williams and Stelfox, 1979). Ascospores are often transported into rapeseed crops from neighbouring cereal crops under which apothecia have formed (Williams and Stelfox, 1979). Apothecia release ascospores continuously for an average of nine days in the field (Schwartz and Steadman, 1978).

Ascospores of Sclerotinia cannot infect leaves and stems directly (Kapoor et al., 1983; Newton and Sequeira, 1972; Purdy and Bardin, 1953). A saprophytic food source provided by petals or other flower parts is necessary for spore germination and plant infection (Lamarque, 1983; McLean, 1958; Natti, 1971). Therefore few, if any, infections are initiated until flowering (Krüger, 1974; Morrall and Dueck, 1982). The period of susceptibility continues as long as petals are present in the crop (Lamarque, 1983). Moist conditions from rainfall or heavy dew are also required for plant infection (Brün et al., 1983; Le Coz, 1981). The most common sites of infection are leaves and leaf axils where fallen petals and water droplets accumulate (Krüger, 1980; Morrall and Dueck, 1982).

The initial symptoms are greyish water-soaked lesions which expand to girdle the stems. Once the stems are girdled,

wilting and premature ripening of the plants occur. Infected stems are brittle, shred easily and have a characteristic bleached appearance. Sclerotia are produced inside infected stems and sometimes externally on infected host tissues. These sclerotia are harvested with the seed or are returned to the soil during harvest, allowing survival of the fungus.

The incidence of stem rot can vary greatly from year to year (Krüger, 1975a; Morrall et al., 1976). Furthermore, disease incidence does not always occur uniformly among fields within a localized geographic region or within an individual field (Gugel and Morrall, 1986; Morrall and Dueck, 1983; Turkington, 1988). Certain aspects of the disease cycle will be discussed in more detail below to explain why these variations exist.

2.1.2 Ascospore production

Sclerotium germination and apothecium formation are influenced by the environment. The optimum temperature range for apothecial production is generally agreed to be 10-20 C (Willettts and Wong, 1980). In Germany, where winter rapeseed is grown, fewer sclerotia germinate and germination is delayed in years with low April soil temperatures (Krüger, 1975a, 1975b). However, in western Canada, the most important factor regulating germination is soil moisture (Morrall and Dueck, 1982). Sclerotia do not germinate under dry conditions. In laboratory studies, sclerotia required water potentials at or

above -7.5 bars in order to produce apothecia (Grogan and Abawi, 1975; Morrall, 1977; Teo and Morrall, 1985). Sclerotium germination is considered a slow process requiring adequate soil moisture for several weeks (Morrall, 1977). However, new apothecia have been observed within two (Boland and Hall, 1987) and four days (Morrall and Dueck, 1983) after rain had rewetted dry soil. Boland and Hall (1987) suggested that sclerotia may be capable of continued carpogenic germination when exposed to a series of short, discontinuous periods of wetting.

A dense plant canopy decreases surface evaporation and dampens soil temperature fluctuations (Abawi and Grogan, 1979; Morrall and Dueck, 1982). Although the development of a dense plant canopy is a requisite for sclerotial germination, sufficient rainfall must occur to produce a favorable microclimate (Morrall and Dueck, 1982).

After sclerotia germinate, humid conditions must be maintained to prevent desiccation of apothecia (Boland and Hall, 1987; Krüger, 1975b). Dehydrated apothecia can be revived and release ascospores when conditions become favorable again (Boland and Hall, 1987; Partyka and Mai, 1962). Heavy rainfall may have a detrimental effect on ascospore release because ascospores disperse into water droplets trapped on apothecia and are washed down into the soil (Krüger, 1974, 1975a, 1975b).

2.1.3 Host infection

The role of petals as an important intermediary in infection by S. sclerotiorum was first demonstrated by Purdy and Bardin (1953) with tomato and is now established with several crops including rapeseed (Abawi et al., 1975; Kapoor et al., 1983; McLean, 1958; Morrall and Dueck, 1982; Natti, 1971). Kapoor et al. (1983) found that ascospores placed directly on rapeseed leaf discs became desiccated and did not germinate. If petal discs were sandwiched between the ascospores and leaf discs, the ascospores germinated and colonization of the petal and leaf discs occurred. Morrall and Dueck (1982) and Lamarque (1983) observed that leaf lesions were often initiated at sites where dead petals were deposited.

Rapeseed petals can become contaminated by ascospores of S. sclerotiorum while still in the inflorescence (Gugel and Morrall, 1986; Lamarque, 1983; Penaud, 1984). Senescent petals are easily dislodged by rain droplets and land on leaves and other plant parts (Penaud, 1984). Excellent retention of petals on leaves occurs under humid conditions, especially if the petals have been colonized by Sclerotinia (Lamarque, 1983; Penaud, 1984). However, excess moisture levels can wash petals off leaves and reduce infection (Lamarque, 1983). Lamarque (1983) obtained the highest frequency of infection when a large number of contaminated petals were clumped together on leaves. She concluded that

the inoculum pressure of S. sclerotiorum provided it with a competitive advantage over other microorganisms.

Ascospore germination and germ tube growth are reported to occur in the absence of free (liquid) water if the relative humidity (RH) is high (Brün et al., 1983; Grogan and Abawi, 1975). However, high RH is difficult to regulate precisely at high temperatures (Rotem, 1988) and thus, periodic condensation may have occurred in these studies. Brün et al. (1983) found that ascospore germination was poor below 88% RH. Ascospores are known to survive as long as 12 days in the field at low RH (Caesar and Pearson, 1983; Grogan and Abawi, 1975). Thus, ascospores can survive a dry period and germinate when conditions become more favorable.

Abawi and Grogan (1975) found that approximately 48-72 hours of continuous leaf wetness were required for infection of bean by ascospores. Infection and lesion expansion did not occur at RH values near 100%. However, Le Coz (1981) successfully infected rapeseed plants under humid greenhouse conditions (>80% RH) without free water. The duration of moist conditions required for infection was inversely related to temperature and ascospore concentration. Brün et al. (1983) suggested that a period of high RH or free water must be maintained for a relatively long period of time, but this period need not be continuous for infection to occur.

In bean, the structure of the plant canopy influences disease development. Canopies that are dense and closed

restrict air movement and produce the most favorable microclimate for infection (Blad et al., 1978; Haas and Bolwyn, 1972; Weiss et al., 1980). A similar situation probably occurs in rapeseed. Turkington (1988) observed that disease incidence was generally higher in crops with dense canopies, especially if lodging had occurred. Lodging accentuates plant to plant spread of S. sclerotiorum (Morrall and Dueck, 1982; Thomas, 1984). Abawi and Grogan (1975) found that white mold of beans was more prevalent in low lying areas and in fields surrounded by wooded areas. Any factor affecting air movement and thus, moisture conditions in the canopy, probably affects the incidence of stem rot.

2.2 Losses due to sclerotinia stem rot

Yield losses due to S. sclerotiorum can be attributed to poorly filled pods, shrivelled seeds and increased shattering during swathing (Krüger and Stoltenberg, 1983; Morrall et al., 1976; Thomas, 1984). Morrall et al. (1976) estimated that yield losses were in the range of 10-15% in heavily infected fields. However, Thomas (1984) stated that losses of over 50% may occur in extreme cases. There are also quality losses. Increased dockage results from shrivelled seeds and sclerotia harvested with the seed (Dueck and Sedun, 1983; Morrall and Dueck, 1983; Thomson and Stelfox, 1983). In addition, sclerotia in seed lots are considered objectionable contaminants in the export market (Petrie and Dueck, 1980;

Verma, 1983). Krüger (1980) and Morrall et al. (1976) compared protein and oil content of seed samples from diseased plants with samples from healthy plants. The disease did not have a major effect on either component.

Morrall et al. (1984a) studied the relationship between yield of rapeseed and stem rot incidence using linear regression analysis. Data collected over a period of several years from fungicide trials and commercial fields were tested. The regression equation $y=a-bx$ (y =actual yield; a =predicted yield without disease; b =constant; x =% of infected plants) was modified so that % yield loss = $100b/a \times (x)$. For regressions with R^2 values greater than 0.6, values of $100b/a$ were generally between 0.4 and 0.5. Thus, if 50% of the plants in a field were diseased the yield loss would be between 20 and 25%.

In later work, R.A.A. Morrall (unpublished) found that values of $100b/a$ varied from 0.34 to 0.76 with a mean of 0.50. He suggested that variations in the values of the multiplier may be related to time of host infection since early infections probably result in the greatest yield reduction. Late infections would be more likely to occur higher up in the crop canopy (because of abscission of lower leaves) causing damage in only a part of the branching inflorescence. Verma (1983) stated that plants infected when fully podded suffer little yield reduction.

Morrall et al. (1976) studied the effects of disease on yield using a different approach. Bundles of plants were collected at random from swaths and the plants were sorted into diseased and healthy categories before threshing. The yield per plant of diseased plants was 22.2 to 54.5% lower than the yield per plant of healthy plants. Percentage reduction in thousand kernel weight closely paralleled percentage reduction in seed yield. Thus, most of the loss in yield was due to a reduction in seed size.

2.3 Chemical control

2.3.1 Fungicides

Commercial use of foliar-applied fungicides to control sclerotinia stem rot in western Canada has become common since 1981 (Morrall et al., 1985). Benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazole carbamate] (trade name Benlate) and iprodione [3-(3,5-dichlorophenyl)-N-(1-methylethyl)-2,4-dioxo-1-imidazoline carboxamide] (trade name Rovral) are currently registered for use in rapeseed in Canada (Morrall et al., 1985; Spencer, 1982). Vinclozolin [3-(3,5-dichlorophenyl)-5-methyl-5-vinylloxazolidine-2,4-dione] (trade name Ronilan) has been used widely in Europe for stem rot control (Davies, 1986; Krüger and Stoltenberg, 1983; Spencer, 1982). Benomyl is a member of the benzimidazole group of fungicides; iprodione and vinclozolin are classified

as dicarboximides (Leroux and Fritz, 1983; Sisler and Ragsdale, 1981). The recommended rate of application is 0.5-0.6 kg active ingredient (a.i.) /ha for all three compounds. Effective control in small plot field trials has also been observed with other fungicides including procymidone, meclozolin and thiophanate-methyl (Dueck et al., 1983; Verma et al., 1983, 1985, 1986, 1987).

A more detailed review of benomyl will aid in the interpretation of results presented in this thesis.

2.3.1.1 Benomyl

Benomyl rapidly decomposes to methyl 2-benzimidazole-carbamate (carbendazim) in aqueous solution and within plants (Clemons and Sisler, 1969; Davidse and Waard, 1984). Carbendazim has been shown to interfere with the formation and functioning of microtubules in fungi (Davidse and Waard, 1984; Sisler and Ragsdale, 1981). This causes disruption of processes such as mitosis and orientation of hyphal growth. Hawthorne and Jarvis (1973) found that benomyl inhibits both ascospore germination and mycelial growth of S. sclerotiorum.

Benomyl has systemic activity (Edgington, 1981; Peterson and Edgington, 1970; Sisler and Ragsdale, 1981). Movement of the breakdown product, carbendazim, is apoplastic ie. through non-living xylem vessels and cell walls (Edgington, 1981; Peterson and Edgington, 1970). Transpiration provides the main driving force for movement within the plant (Davidse and

Waard, 1984). Therefore, translocation of carbendazim is mainly upward into organs that have an ability to transpire eg. leaves (Davidse and Waard, 1984; Peterson and Edgington, 1971). Research has shown that systemic movement into blossoms is minimal (Hunter et al., 1978; Peterson and Edgington, 1971). Peterson and Edgington (1970) found that movement in bean leaves was acropetal. Carbendazim became concentrated at the margins and apices of leaflets. Similarly, Hunter et al. (1978) observed that benomyl applied to the base of a bean leaf protected the entire leaf against S. sclerotiorum, but benomyl applied to the leaf tip did not protect the base.

Rapeseed petals, leaf bases and petioles are plant structures commonly involved in the disease cycle of stem rot (Section 2.1). However, accumulation of chemical in these plant parts through systemic movement probably does not occur. Thorough plant coverage especially of blossoms, may be more important than systemic movement for good control (Steadman, 1979; Willetts and Wong, 1980). Hunter et al. (1978) achieved effective control of S. sclerotiorum on bean when only the blossoms were sprayed with benomyl. A single application of benomyl can protect petals against colonization by Sclerotinia for several days (Natti, 1971; Reynaud, 1983). Reynaud (1983) found that germination of ascospores on senescent rapeseed petals was inhibited up to 13 days after treatment.

Benomyl is effective as a preventive spray before infection and has little curative action (Hunter et al., 1978; Regnault and Pierre, 1984). In greenhouse tests, Hunter et al. (1978) found that benomyl sprays applied to bean plants 72 hours after inoculation with ascospores were completely ineffective. Therefore, delaying treatment until stem rot symptoms appear will not result in effective control.

2.3.1.2 Fungicide resistance

Continuous and widespread use of a fungicide or group of fungicides may lead to the development of resistant strains in the pathogen population (Agrios, 1978; Sisler and Ragsdale, 1981). Resistance to benomyl has been reported in several fungi including Sclerotinia minor, a species closely related to S. sclerotiorum (Davidse and Waard, 1984; Willetts and Wong, 1980). Strains of S. sclerotiorum resistant to dicarboximide fungicides have been isolated in the laboratory (Leroux and Fritz, 1983).

2.3.2 Fungicide efficacy

Foliar sprays have been used effectively in many crops including bean, sunflower, lettuce, cabbage and tomato to control diseases caused by S. sclerotiorum (Steadman, 1979; Willetts and Wong, 1980). Several experiments in Europe and western Canada have shown that benefits can result from

chemical control in rapeseed (Davies, 1986; Dueck et al., 1983; Morrall et al., 1985; Thomson et al., 1984).

Dueck et al. (1983) evaluated fungicide application using small plot and large scale field tests in 1978 and 1980. Tests were conducted in naturally infested commercial rapeseed fields (Brassica napus cultivars Regent and Midas; B. campestris cultivar Candle) in Saskatchewan and Alberta. Application was made when the plants were at approximately 25% bloom (early bloom). In small plots, treatments included benomyl, iprodione and vinclozolin applied at 0.25-1.0 kg a.i./ha. All three fungicides reduced stem rot significantly in 1978. In 1980 under higher disease pressure, benomyl and vinclozolin significantly reduced disease but iprodione did not. Better disease control was achieved at higher rates of application for benomyl and vinclozolin. Yield differences among treatments were difficult to detect due to stand variation and seed shrivelling from premature harvesting. In large scale tests, aerial application of benomyl (1.0 kg a.i./ha) and iprodione (0.5 kg a.i./ha) provided control equivalent to comparable treatments in small plot tests (Dueck et al., 1983). In 1980 benomyl- and iprodione-treated strips yielded significantly more than check strips and were not significantly different from each other.

Fungicide trials were carried out in Alberta by Thomson et al. (1984) in 1981 and 1982. In 1981 6 of 10 fields were planted to the cultivar Altex (Brassica napus) and the others

were planted to Candle (*B. campestris*). The fungicides tested were benomyl (0.55 kg a.i./ha) and iprodione (0.75 kg a.i./ha). In 1982 one field of Altex and one field of Candle were sprayed with benomyl at 0.50 kg a.i./ha. In both years spraying was done by airplane at the early bloom stage. Benomyl significantly reduced disease in both cultivars in 1981, and significantly increased yield in Altex but not in Candle. The yield of Candle was substantially increased by fungicide application only in 1982 when disease levels were higher than in 1981. Iprodione gave results similar to those with benomyl. Thomson et al. (1984) found that fungicide application also resulted in fewer sclerotia with the seed and reduced dockage levels.

In Germany Krüger and Stoltenberg (1983) studied the effect of vinclozolin application on yield of winter rapeseed using data assembled from numerous experiments. They found that there was a significant low correlation ($r=0.48$) between the relative yield increase from spraying and disease incidence. At low disease incidence (0-12%) spraying resulted in average yield increases of 2-5%. At infection levels of 13-40% average yield increases were 9-14%. Yield increases averaged 26% in fields with high infection levels (40%).

The above and other studies (Morrall et al., 1983, 1984b; Verma et al., 1983, 1985, 1986, 1987) have shown that effective control of stem rot can be achieved by foliar

application of fungicides. However, treatments have not always resulted in significant yield increases.

2.3.3 Factors affecting efficacy of foliar fungicide application

2.3.3.1 Timing

The best time to apply fungicides for the control of Sclerotinia diseases with ascosporic inoculum is during flowering, before infections are initiated (Krüger and Stoltenberg, 1983; Natti, 1971; Steadman, 1979). Timely blossom coverage to prevent colonization of senescent flower parts is essential for good control (Steadman, 1979).

Aerial application of fungicide to control sclerotinia stem rot of rapeseed was most effective when the crop was in early bloom (Dueck et al., 1983; Thomas, 1984). Lack of control at later stages of bloom was attributed to failure of the fungicide to penetrate the denser crop canopy. Thomas (1984) stated that an aerial fungicide application may not be economical if there are more than four pods set on the main stem of the majority of plants. Effective control can be achieved with ground application at either early, or full bloom (Morrall et al., 1984b, 1985). Morrall et al. (1984b) obtained significant disease reductions in three test fields after application of benomyl, iprodione, or vinclozolin at full bloom using a commercial 'spra-coupe'.

Fungicide application at late bloom has usually resulted in poor stem rot control (Morrall et al., 1983, 1984b; Verma et al., 1983). According to Krüger (1973), spraying should more or less coincide with ascospore release. In the studies involving late bloom application, apothecia were present in the test fields (or adjacent fields) when the crops were in early bloom. Ascospores were probably widespread in the crops two or three weeks before the fungicide application. Thus, many infections may have been well advanced by late bloom and this would have limited control.

2.3.3.2 Ground versus aerial application

Aerial and ground application for control of sclerotinia stem rot in rapeseed in western Canada were compared by Morrall et al. (1983). Benomyl at 0.6 kg a.i./ha was applied with a tractor mounted sprayer using 124 l water/ha or by airplane using 42 l water/ha. Different dates of application were tested in each of four fields. Ground application gave significantly better disease control than aerial on at least one treatment date in three of four fields. Morrall et al. (1983) attributed the better control to better penetration of the canopy, presumably due to higher water volume and the position of the spray nozzles. Yield loss from tractor wheel damage was estimated at 4% when spraying was done at the full bloom stage. Estimated costs of commercial spraying (excluding fungicide) were \$8.65/ha for tractor application

and \$11.10/ha for aerial application (Morrall et al., 1983).

Morrall et al. (1985) suggested three advantages of ground over aerial application: " a) superior disease control, b) a greater 'window' when spraying is likely to be effective; by extending the effective period from early through to full bloom, the grower has more time to make a decision about spraying and to act, c) lower costs of application."

In western Canada, most commercial fungicide application to date has been by fixed wing aircraft (Morrall et al., 1985). Equipment for ground spraying has been used by some growers for several years now. According to Morrall et al. (1984b), a commercial 'spra-coupe' can be driven through rapeseed at full bloom with minimal crop damage at a speed of 14 kph. The 'spra-coupe' could be used to spray 12 ha/h, including the time for tank mixing.

2.3.3.3 Rate of chemical

In 1980 benomyl was ground-applied at 0.25, 0.50 (the recommended rate), 0.75 and 1.0 kg a.i./ha in small plot field tests (Dueck et al., 1983). In this study disease pressure was high. Although better disease control was achieved as the rate of application was increased, significant disease reductions were obtained using 0.25 kg a.i./ha. Applications at 0.25 kg a.i./ha reduced disease severity to about half that in the checks. The data obtained by Dueck et al. (1983)

suggested that reduced rates of fungicide application warranted further investigation (Morrall et al., 1985).

Morrall et al. (1984b) applied benomyl, iprodione or vinclozolin at 0.3 kg a.i./ha at early bloom using a commercial 'spra-coupe'. Dual applications at early and full bloom, each at 0.3 kg a.i./ha, were also tested. The single half-rate applications at early bloom often reduced disease significantly and gave good control at low disease pressure. The split applications gave excellent control even at high disease pressure. Iprodione was generally less effective than the other two fungicides. Morrall and Verma (1988) found that iprodione reduced disease significantly when applied at 0.5 kg a.i./ha but at lower rates control was not effective. Recent research has shown that benomyl can significantly lower disease incidence at rates as low as 0.125 kg a.i./ha (Morrall, 1988; Morrall and Verma, 1987).

According to Morrall et al. (1985), when weather immediately before bloom is only marginally favorable for disease development, growers could spray at half the recommended rate at early bloom and be assured of at least some disease protection. If the weather subsequently turned wetter, a second half-rate application could be made at full bloom to increase protection. These options depend upon the use of ground rigs for application (Morrall et al., 1985).

2.3.3.4 Physical aspects of application

Rogers et al. (1988) studied the effect of droplet size on deposit efficiency on rapeseed by adding a fluorescent dye to a benomyl spray solution. Nozzle types were chosen to give droplet sizes ranging from ultra small droplets (130 micron diameter) to conventional droplets (410 micron diameter). The workers found that reduced droplet size significantly increased deposit efficiency on five plant parts. Throughout the plant the deposit from ultra small droplets was approximately 2.5 times that from conventional droplets.

There is some evidence that nozzles generating small droplets provide superior stem rot control. Morrall and Verma (1987) found that disease incidence was lower in treatments applied with 11001 nozzles compared to those applied with 80015 nozzles (reported as 8015 nozzles). Morrall (1988) obtained excellent control under low disease pressure using 800017 nozzles (ultra small droplets) with 552 kPa pressure and only 10 l carrier fluid/ha. If small droplets are used, high carrier volumes may not be required for thorough crop coverage and good disease control. Unfortunately, small droplets are also more prone to wind drift (Spillman, 1984).

2.3.4 Economic considerations

In western Canada, the use of either benomyl or iprodione at the recommended rates costs about \$50 CAN/ha for chemical and application. The high cost of spraying makes disease

control economically questionable if the yield potential of the crop is poor. The market value of rapeseed affects the economics of chemical control. With an average price of \$0.26/kg for rapeseed, a yield increase of at least 190 kg/ha is needed to make spraying economically justifiable. Growers with good management practices commonly achieve yields ranging from 1600-2300 kg/ha (Thomas, 1984). Thus, a yield increase of 8-12% would be required to recover spraying costs in western Canada. However, in years with low rapeseed prices (eg. 1986) yield increases of 12-20% are necessary (R.A.A. Morrall, personal communication).

Disease levels of 15-30% infected plants or more cause yield losses high enough to justify fungicide application (Krüger and Stoltenberg, 1983; Morrall et al., 1976; Thomson et al., 1984). In western Canada, disease incidence values in rapeseed fields are generally less than 20% (Gugel and Morrall, 1986; Morrall and Dueck, 1982; Turkington, 1988). Therefore, in most fields chemical control is not warranted. Disease forecasting methods have been developed to help growers identify fields that require fungicide application (Thomas, 1984; Turkington, 1988). Unfortunately, these methods have not always been reliable. Better disease forecasting combined with new options such as half-rate and split fungicide applications may help lessen the financial risks associated with chemical control.

2.4 Forecasting stem rot epidemics in western Canada

The decision by a rapeseed grower on whether to spray or not has to be made before symptoms of stem rot become visible. Because of the variable incidence of stem rot, disease forecasting is important for rational fungicide use. Growers can benefit from accurate disease prediction by: a) avoiding unnecessary fungicide application when a low disease risk is predicted, and b) minimizing disease losses by spraying a fungicide when a high risk is predicted.

2.4.1 Host-pathogen-weather based forecasting system

A qualitative disease forecasting system has been developed in western Canada to help growers decide if spraying a field is required (Thomas, 1984). The system consists of a checklist with points assigned for the answers to questions based on host, pathogen and weather factors. Considerations include: disease history of the field and area, previous and current weather conditions, presence of apothecia, stand density and potential yield. If the total score exceeds a predetermined level then economic returns from spraying are probable.

This forecasting method has a few limitations. Searching for apothecia is difficult and often unreliable (Kaminski, 1987). Apothecia are small, inconspicuous and nonuniformly distributed and thus, can easily escape detection. In

addition, this system does not adequately account for ascospores that move into the crop from extrinsic sources (Gugel, 1985; Gugel and Morrall, 1986). According to Gugel (1985), the accuracy of stem rot forecasts could be improved if a clear quantitative relationship between inoculum density and final disease levels were demonstrated.

2.4.2 Forecasting based on ascospore infestation of petals

Gugel (1985) conducted a series of experiments to determine if the frequency of ascospore contamination of various plant structures was correlated with disease incidence. Live and dead petals, leaf axils and leaf bases were collected at sites in commercial rapeseed fields in Saskatchewan and tested for the presence of ascospores by plating on a selective agar medium. Disease incidence was assessed at the same sites before swathing in August. Gugel (1985) found that a significant relationship existed between infestation of live petals at early bloom and final disease. He concluded that this relationship could possibly be exploited in a disease forecasting system.

Turkington (1988) investigated the feasibility of using frequency of ascospore infested rapeseed petals in a commercial stem rot forecasting system. A pilot project was set up whereby cooperating growers collected rapeseed petals during early bloom and brought them in to a laboratory in

east-central Saskatchewan. There the percentage petal infestation was determined by trained technicians using an agar plate test. Petal test results and a subsequent estimate of disease risk (low, moderate, or high) were given to growers three to five days after petal collection. In crops that had been sampled disease incidence was determined before swathing. Turkington (1988) found that forecasts based on petal tests were relatively accurate in identifying crops at low disease risk. However, forecasts were not as reliable for crops at moderate and high disease risk. This was attributed to changing moisture conditions and inoculum levels after sampling. Turkington (1988) suggested that repeated petal sampling during flowering might result in more accurate forecasts. Canopy density was an important factor affecting the petal infestation-disease incidence relationship. Turkington (1988) concluded that petal testing, if used in conjunction with qualitative criteria (eg. canopy density), should provide a reliable and easy-to-use forecasting system.

Kaminski (1987) assessed the suitability of a petal testing kit for home use by rapeseed growers in Saskatchewan. The kit contained necessary materials (eg. disinfectant, culture plates) and instructions for setting up a petal test. Growers and trained technicians collected petal samples from the same sites in rapeseed fields at about the same time. Both sets of individuals carried out the petal testing procedure independently. Identification of S. sclerotiorum

on culture plates was always left to trained personnel. Kaminski (1987) found that estimates of percentage petal infestation based on tests set up by growers were not significantly different from those based on tests done by trained individuals. Unfortunately, a comparison could not be made over the full range of possible values of percentage petal infestation. Most crops had very low values because of abnormally dry conditions in June. Kaminski (1987) concluded that the kit was suitable for home use by growers. He thought that a commercial product based on the prototype kit could be marketed in the future.

3. Flowering Stages in Rapeseed

In the present study the flowering stages first flower, early bloom, full bloom and late bloom were differentiated (Table 3-1). These growth stages approximate stages 4.1 to 4.4 in the Harper and Berkenkamp (1975) key.

Table 3-1 Description of flowering stages in rapeseed.

Stage	Description
First flower	First flowers open
Early bloom	10-15 flowers open on main stem, lower leaves still attached
Full bloom	Many flowers open on main stem and side branches, lower pods elongating, leaf abscission starting, older petals fallen
Late bloom	Few unopened buds, lower pods starting to fill

The flowering period in western Canada can last 21 days or longer, depending on environmental conditions. The flowering period of Brassica napus cultivars is longer than that of B. campestris cultivars. Late bloom is a relatively long stage, especially when moist conditions prolong flowering.

4. Field Studies to Evaluate the Efficacy of Fungicide Application at Late Bloom

4.1 Introduction

The objectives of these experiments were twofold:

a) to evaluate the efficacy of fungicide application at late bloom. Nozzle type and rate of application were also varied in some trials.

b) to determine if the flowering stage at which Sclerotinia ascospores first become widespread in the crop is a factor affecting the efficacy of late bloom application.

4.2 1987

4.2.1 Experiment dependent on artificial inoculation

4.2.1.1 Materials and methods

A factorial experiment in a split-plot RCBD design was set up at Saskatoon and at Canwood (180 km N of Saskatoon). The rates of benomyl applied at late bloom were the subfactor. The rates tested were 0 (check), 0.3 and 0.6 kg a.i./ha. Three periods of inoculation corresponding to early, full and late bloom and an uninoculated check were the main factor. Five replications were used at each location.

4.2.1.1.1 Saskatoon

The test site was treated with the herbicide Treflan QR5 (5% trifluralin) at a rate of 25 kg product/ha before seeding. Plots were seeded to Brassica napus cv. Westar on May 12 at a rate of 6 kg/ha with a small-plot tractor-drawn seeder. Each sub plot was 4.9 x 2.5m. Strips of barley 1.3m wide were seeded between replicates and between main plots within a replicate in order to provide pathways and minimize wind drift of applied inoculum. To control flea beetle (Phyllotreta cruciferae) the insecticide Furadan (carbofuran) was applied with the seed and the insecticide malathion was applied as a foliar spray when the plants were in the rosette stage. Drought stress was prevented by watering the plants using a sprinkler irrigation system. Periodic hand weeding ensured weed control.

Inoculum was introduced by spraying a suspension of ascospores and hyphal fragments on the plants using a backpack sprayer with a hand-held boom. Ascospore suspensions were produced by grinding approximately 30 mature apothecia in distilled water using a mortar and pestle. Suspensions containing hyphal fragments were produced by blending mycelium of S. sclerotiorum grown on Difco potato dextrose agar (PDA) using a Waring blender. The ascospore and hyphal suspensions were mixed and filtered through cheesecloth. Inoculum suspensions were applied on two different dates for each period of inoculation. Dates of inoculation were June 26 and

29, July 1 and 5, and July 7 and 9 for early, full and late bloom-inoculated plots respectively. If the plant canopy was dry, irrigation water was applied before inoculation to enhance infection. Throughout the bloom period the plants were frequently watered lightly in an attempt to maintain water droplets in the canopy.

On June 30 10 plant tops were collected from the plots that had been inoculated on June 26 and 29 at early bloom. Forty petals were picked from the inflorescences and plated 4 per plate on sterile Difco PDA amended with 25 ppm anhydrous ampicillin and 25 ppm streptomycin sulfate. Turkington (1988) found that this medium permits growth of S. sclerotiorum while assuring good protection from bacterial contamination. The plates were incubated at room temperature for four days and then each petal was scored for the presence of Sclerotinia. The purpose of this test was to confirm that viable inoculum was present on the petals of inoculated plants.

Applications of benomyl were made with a backpack sprayer operated at 240 kPa with 8003 nozzles and using 900 ml of water/plot. The chemical was applied on July 10.

In early August the test was abandoned because stem rot symptoms were not observed in any plots.

4.2.1.1.2 Canwood

The plots were established in a commercial crop of B. napus cv. Westar when the plants were in the early bud stage. In late June the plants were wilting from drought stress. On June 27, when the crop was in early bloom, water was hauled to the test site in a 4500L container. Water was applied approximately equal to 13mm of rainfall.

Plot size and method of chemical application were similar to the Saskatoon test. There were no strips of barley but subplots were separated by 1.3m guard strips. The method of inoculation differed in that the inoculum suspension contained ascospores but not hyphal fragments. Dates of inoculation were June 27 and 29, July 2 and 4, and July 11 and 13 for early, full and late bloom-inoculated plots respectively. Applications of benomyl were made on July 11.

Disease ratings were made on August 12-14 when the plants were nearly mature. About 200 plants were pulled from the center of each plot and classified into 5 categories using a rating scale (Appendix A). A disease index was calculated using the following formula:

$$\text{Index} = \frac{\sum (\text{No. of plants in a category} \times \text{category value}) \times 100}{\text{Total no. of plants} \times \text{max. category value}}$$

Disease incidence was obtained for each plot by calculating the percentage of plants with stem rot symptoms without regard to category.

A rain gauge was placed in a farmer's yard approximately 2 km from the test site. Rainfall data were obtained for June and July.

4.2.1.2 Results and discussion

4.2.1.2.1 Saskatoon

No disease developed despite a dense plant canopy. The frequency of petals infested with S. sclerotiorum on June 30 was 55%. Therefore, the suspensions applied to the plants contained viable inoculum. The absence of disease was probably due to abnormally hot, dry weather that persisted for much of the flowering period. Environmental data for the Saskatoon area obtained from the Saskatchewan Research Council climatological reference station revealed that the mean daily temperature in June (18.9C) was 3.3C above normal (15.6C). Total precipitation for June was 22.0mm, well below the normal (59.9mm). July was also a warm and dry month with near normal temperatures and below normal precipitation. Despite frequent irrigation during flowering there was probably never a sufficiently long period of leaf wetness and/or high RH for infections to occur. Irrigation may have even reduced the likelihood of disease by washing inoculum off the plants.

4.2.1.2.2 Canwood

The arcsine transformation ($\arcsin \sqrt{\text{percentage}}$) was applied to both disease incidence and disease index data before analyses. This was necessary because most values were in the range of 0-20%. Arcsine transformation of such data ensures that variance is not related to the mean, one of the assumptions of the analysis of variance (Sokal and Rohlf, 1981). The data were subjected to analysis of variance and, when the F ratio was significant, means were separated using an LSD test. The same conclusions were drawn by analysing either index or incidence data. Therefore, only analyses of incidence data are presented.

Significant differences among inoculation treatments and rates of benomyl application were detected (Table 4-1). The interaction was not significant.

Application of benomyl at late bloom significantly reduced disease incidence (Table 4-2). Effective control was achieved by spraying at 0.3 kg a.i./ha, half the recommended rate. There was no significant difference in control between half-rate and recommended rate applications.

Significantly more disease developed in early bloom inoculated plots than in full and late bloom inoculated plots (Table 4-3). This is consistent with results reported by Gugel (1985). He obtained stronger relationships between inoculum and disease at early rather than full or late bloom. Rapeseed plants begin shedding leaves at full bloom. Thus,

Table 4-1 Analysis of variance of incidence of stem rot in relation to 4 inoculation treatments and 3 rates of benomyl application for the Canwood experiment in 1987.

Source	df	MS	F
Block	4	149.7	
Inoculation	3	372.3	10.0*
Error (a)	12	37.1	
Rate of application	2	1161.4	57.8*
Inoculation*rate interaction	6	6.3	<1
Error (b)	32	20.1	

* Significant at $p=0.05$.

Table 4-2 The effect of benomyl application at late bloom on incidence of stem rot in the Canwood experiment in 1987.

Rate of benomyl application (kg a.i./ha)	Disease incidence ⁺
0 (check)	15.7a [^]
0.3	4.3b
0.6	2.8b
SE 1.2	

+ All figures represent means of 20 observations.

[^] Values followed by same letter do not differ significantly from one another according to a protected LSD test at $p=0.05$ after arcsine transformation of the data.
SE = standard error.

infections initiated in leaf tissues at full or late bloom may not progress into the main stem before abscission occurs. Even under favorable environmental conditions high levels of

petal infestation at late bloom may not be associated with high disease incidence.

Some natural inoculum was present because stem rot was evident in plots that were not inoculated and in the crop surrounding the test site. The mean disease incidence was 10.0 in plots that were not inoculated and not treated with fungicide (Figure 4-1). Surprisingly, plots that were not inoculated showed more disease than plots inoculated at late bloom although the difference was not significant (Table 4-3).

Benomyl application was approximately equally effective under all 4 inoculation treatments (Figure 4-1). This accounts for the nonsignificant interaction term in the analysis of variance (Table 4-1). Benomyl has little curative

Table 4-3 The effect of period of inoculation on incidence of stem rot in the Canwood experiment in 1987.

Inoculation treatment	Disease incidence ⁺
early bloom	12.9a [^]
full bloom	8.9b
no inoculation	4.6bc
late bloom	4.0c
SE 1.3	

+ All figures represent means of 15 observations.

[^] Values followed by the same letter do not differ significantly from one another according to a protected LSD test at $p=0.05$ after arcsine transformation of the data.

SE = standard error.

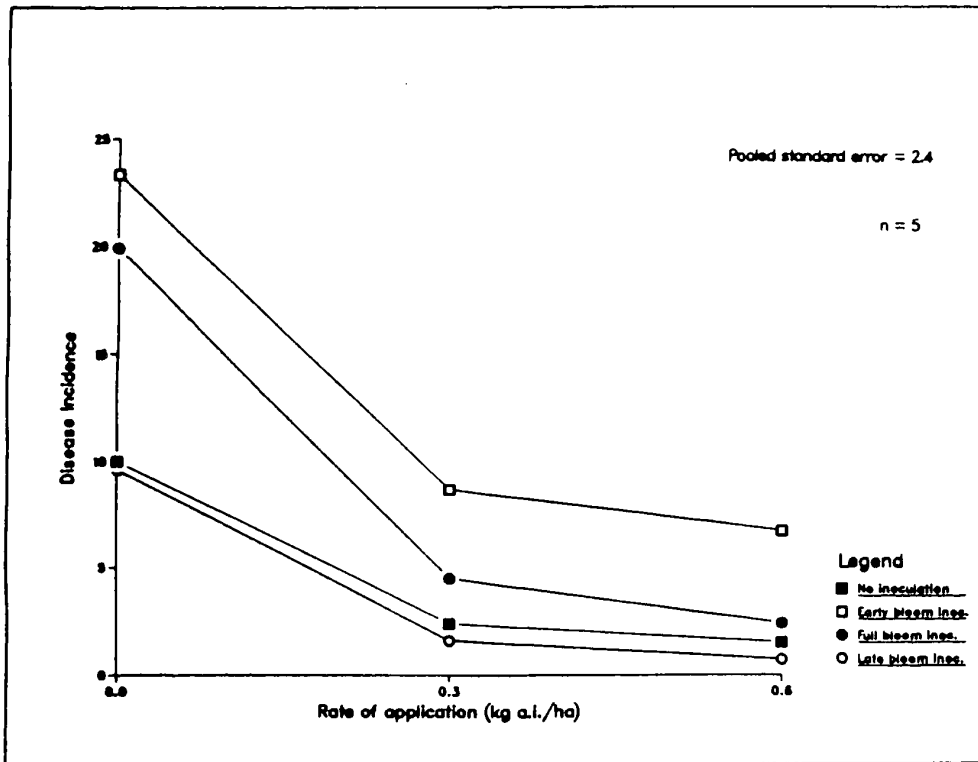


Figure 4-1 The effect of benomyl application at late bloom on incidence of stem rot in relation to period of inoculation, Canwood 1987.

action (Section 2.3). It was expected that many infections in the early bloom-inoculated plots would have been well advanced by late bloom and this would have limited the efficacy of the fungicide. However, significant control was achieved even in those plots inoculated at early bloom.

Weather conditions were dry in Canwood for most of June (Table 4-4). It is probable that very few, if any, apothecia were present in the area in June. Sclerotia probably germinated in response to extended moist soil conditions resulting from the June 29-July 1 rainfall. Ascospores were

probably not widespread in the area until mid-July. Thus, infections caused by 'background inoculum' would have been initiated fairly late in the bloom period. This might partially account for the better than expected control in the early bloom-inoculated plots. Many ascospores applied to plots at early bloom may not have germinated and infected plants until late bloom. Frequent showers occurred from July 10-14 (Table 4-4), providing ideal infection conditions during late bloom. This may have also contributed to the unexpected results.

Table 4-4 Rainfall data obtained for June and July using a commercial rain gauge, Canwood 1987.

Date(s)	Rainfall (mm)	Date(s)	Rainfall (mm)
June 1-9	0	July 1	10.2
10	7.6	2-3	0
11-15	0	4	10.2
16	5.1	5	7.6
17-19	0	6-9	0
20	12.7	10-13	7.6
21-28	0	14	30.5
29	10.2	15-18	0
30	27.9	19	22.9
		20	5.1
		21-31	0
June Total	63.5	July Total	94.1

4.2.2 Experiment dependent on natural infection

4.2.2.1 Materials and methods

The experiment was conducted in three commercial crops of B. napus cv. Westar with moderate to heavy stands in the Melfort area (200 km NE of Saskatoon). In field 87-1, apothecia were first observed when the crop was in transition between full and late bloom. An agar plate test (see Section 4.2.1.1.1) at late bloom confirmed the presence of S. sclerotiorum on the petals. In field 87-2, apothecia were first observed when the crop was in full bloom. Petal tests indicated that inoculum was present at full and late bloom but not at early bloom. Abundant apothecia were present from the late-bud stage onward in field 87-3. Petals were infested with Sclerotinia at early, full and late bloom.

The trial at each location was divided into 24 plots (2m x 5m). Twelve plots were sprayed with benomyl (0.6 kg a.i./ha) at late bloom. The plots were paired so that a sprayed plot was always adjacent to a check plot. Treatments were applied with a hand-held applicator at 400 l/ha and 240 kPa using LF3 80 nozzles. Spray dates were July 9 for field 87-1 and July 17 for fields 87-2 and 87-3.

The trials were assessed for disease when the plants were nearly mature (August 17 for fields 87-1 and 87-2, August 19 for field 87-3). In fields 87-1 and 87-2, plants were pulled from a 2m² area in the center of each plot. A 3m² area was

used in field 87-3 because of lower plant density. Disease ratings were performed and disease incidence and disease index values were calculated for each plot as described previously for the Canwood experiment. Yield data were obtained in fields 87-2 and 87-3. The plants from the harvested area were placed in flour bags and taken to Saskatoon for drying and later combining. Final seed yields were determined after cleaning and weighing the seed.

4.2.2.2 Results and discussion

The data were analysed using paired t-tests. The arcsine transformation was applied to incidence and index data before analyses. Since analyses of index data provided no additional insights over analyses of incidence data, only incidence data are presented here.

Application of benomyl at the recommended rate at late bloom significantly reduced the amount of stem rot in all three fields (Table 4-5). A significant yield increase from spraying did not occur in either field 87-2 or 87-3. The yield in check plots was in fact higher than the yield in sprayed plots at both locations although the differences were not significant. Yield differences may have been masked because of seed shrivelling due to slightly premature harvesting and variations in stand density. Yield differences among treatments are often difficult to determine in small plot experiments, especially if disease levels are fairly low (Dueck et al., 1983).

Table 4-5 The effect of benomyl application at late bloom on incidence of stem rot and yield in three rapeseed fields, Melfort 1987.

		Field 87-1	Field 87-2	Field 87-3
Disease incidence (SD)	sprayed	1.7 ⁺ (0.3)	4.2 ⁺ (0.8)	22.2 ⁺ (4.6)
	check	16.8 (3.6)	24.5 (2.1)	36.4 (6.0)
Percentage control [^]		90	83	39
Yield in g (SD)	sprayed	--	365(10)	414(127)
	check	--	380(22)	456(138)

All figures represent means of twelve replications.

+ Significantly different from check at p=0.01 (paired t-test after arcsine transformation of the data).

[^] $\frac{\text{check} - \text{sprayed}}{\text{check}} \times 100$

SD = standard deviation.

Excellent control was achieved in fields 87-1 and 87-2. Ascospores of S. sclerotiorum were not present until the full bloom stage or later in these fields because dry weather in June had delayed germination of sclerotia. Thus, the fungicide was probably applied at approximately the same time that infections were initiated. In field 87-3 inoculum was present at the onset of flowering because of frequent showers in June. Many infections were probably well developed by late bloom and, as a result, fungicide application at late bloom was much less effective than in the other two fields.

4.3 1988

In 1988, all experiments were conducted in commercial fields and were dependent solely on natural infection. This provided several advantages over artificial inoculation. Firstly, the labor intensive procedures of preparing and applying the inoculum were eliminated. The savings in time provided by relying on natural infection allowed more tests to be conducted, thereby increasing the likelihood of an epidemic of stem rot occurring in at least one of the tests. Secondly, since 'background inoculum' would be the sole source of inoculum, interpretation of results would be simplified. Finally, rainfall soon after artificial application of inoculum would tend to wash the inoculum off petals and, hence, might reduce the final disease incidence. This would not be a problem in experiments depending on natural inoculum which would be deposited on petals over a prolonged period.

4.3.1 Selection of test sites

Though originally planned for the Melfort area, studies were conducted near Meadow Lake, Saskatchewan (250 km NW of Saskatoon) because of abnormally dry weather at Melfort in June. The Meadow Lake area was chosen because a) it has a history of stem rot and b) moisture conditions in June and early July were conducive to good crop growth, inoculum production and disease development.

Rapeseed crops with moderate or heavy stands were located in the last week of June and first week of July. The frequency of petals infested with S. sclerotiorum was monitored in 27 of these crops throughout the bloom period. Typically, petal samples were collected from five widely spaced sites (>50m apart) in each crop. At each site 10-15 plant tops were picked and placed in a clean plastic bag. The plant growth stage was recorded and the samples were taken to an office in Meadow Lake where they were processed within 3 hours. Petals were picked and plated on agar medium as described in Section 4.2.1.1.1. Forty petals were plated per site. After 4 days incubation the mean percentage of petals infested per site and per crop was determined. Each crop was sampled every 2-5 days beginning at first flower (or slightly later with some crops) and continuing until flowering was almost complete.

By monitoring petal infestation, tests could be established in crops where abundant inoculum was present at early bloom, as well as in crops where the appearance of inoculum was delayed until full or late bloom. The petal infestation data were combined with observations on stand density, uniformity and weediness in choosing the most desirable fields for test sites.

4.3.2 Experiment to evaluate different rates of application and different nozzle types

4.3.2.1 Materials and methods

The experiment was set up at six locations. Fields 88-1, 88-2, 88-4, 88-5 and 88-6 were planted to Brassica napus cv. Westar and field 88-3 was planted to B. campestris cv. Tobin. The plots were established and benomyl was applied when the crops were in the late bloom stage. Treatments consisted of a factorial combination of two nozzle types (LF3 80 , 11001) and four rates of application (0, 0.15, 0.3, and 0.6 kg a.i./ha). Plots were 5m x 2m. The plot design was a randomized complete block with four replications. Treatments were applied with a hand-held applicator at 240 kPa with 400L H₂O/ha. In field 88-3, the chemical was not applied until the crop had almost finished flowering because adverse weather conditions (wind, heavy rain) delayed spraying. Spray dates were July 7 for fields 88-3 and 88-5, July 8 for field 88-6, July 9 for fields 88-2 and 88-4 and July 18 for field 88-1.

Plots were assessed for disease in August when the plants were nearly mature. Disease incidence was determined by counting the percentage of infected plants in a sample of 200 plants in the center of each plot. In field 88-4, disease severity was also assessed by classifying individual plants using a rating scale and determining disease indices as in Section 4.2.1.1.2. All data from fields 88-1, 88-3 and 88-4

were arcsine transformed and subjected to analysis of variance. Means for rate of application were separated using protected LSD tests. The two nozzle types were compared with an F test after excluding the data from check plots. Disease severity data was also collected in the check plots in fields 88-1 and 88-3 for use in the study of I-S relationships (Section 6).

4.3.2.2 Results and discussion

4.3.2.2.1 Percentage petal infestation

The percentage petal infestation (PPI) during the bloom period for all six fields is shown in Figure 4-2. For each field only the PPI data for the sampling site closest to the fungicide trial were plotted. Means in each field based on all five sites were also calculated (Appendix B).

In fields 88-1 and 88-4, inoculum was present on the petals beginning at early bloom (Figure 4-2). In fields 88-2, 88-5 and 88-6, the PPI was very low at early bloom but increased to 30-50 by full bloom. Petal samples were not taken in field 88-3 until full bloom. Inoculum levels were probably very low at early bloom because weather conditions during late bud and early flowering were not conducive to sclerotium germination. A substantial decrease in PPI occurred in fields 88-1 and 88-3 on the first late bloom

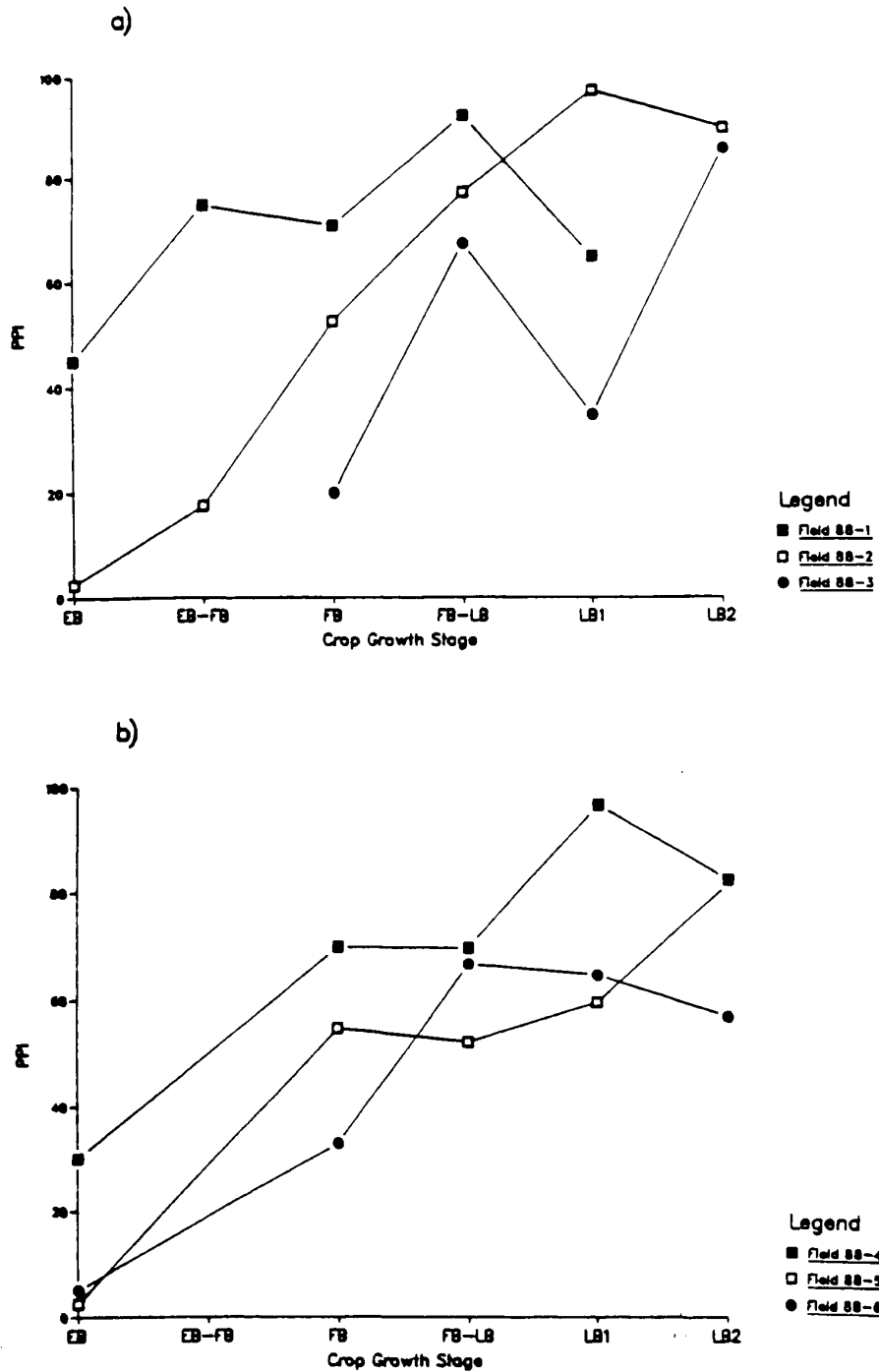


Figure 4-2 Percentage petal infestation (PPI) during the bloom period for six crops, Meadow Lake 1988 (EB = early bloom, FB = full bloom, LB = late bloom).

a) Fields 88-(1-3) b) Fields 88-(4-6)

sampling date (LB1). However, since the petal samples had been collected soon after a heavy rain, some inoculum had probably been washed off the petals. The PPI reached 60 or greater by late bloom in all six fields. Thus, all six crops had the potential for considerable disease development.

4.3.2.2.2 Variations in disease levels among tests

The mean disease incidence in the check plots was less than 2% in fields 88-(2,5 and 6). Because disease levels were very low the data from these tests were not analysed.

When the crops in fields 88-(2,5 and 6) were in early and full bloom (June 28-July 7), environmental conditions at Meadow Lake were favorable for ascospore germination and host infection (Appendix C). Rainfall was recorded on 8 of 10 days during this period. However, ascospores were not widespread in these fields until approximately July 1 when the crops were in full bloom (Appendix B). Infections were probably initiated in leaf tissues during the period July 1-7. However, because disease levels were very low, it is possible that the pathogen did not have time to progress into the main stem before abscission of infected leaves occurred. The weather was warm and dry during mid- and late-July (Appendix C). From July 15 to 29 only 6.2mm of rainfall was recorded. The maximum daily temperature during this period ranged from

21-31C. These conditions were not conducive to the development of 'late infections'.

Disease did develop at the other three locations. In fields 88-1 and 88-4 the mean disease incidence in the check plots was 18.0 and 44.6 respectively. Inoculum was abundant in these two fields when the crops were in early bloom (Figure 4-2, Appendix B). Many infections were probably initiated at early bloom because of moist conditions at this time (early July). Leaf abscission in rapeseed does not begin until full bloom. Thus, infections initiated at early bloom have a longer period for establishment in the main stem than infections initiated later in the bloom period. This might explain why disease developed in these two fields but not in fields 88-(2,5 and 6).

In field 88-3, the mean disease incidence in the check plots was 28.6. This field was planted to the early maturing B. campestris cv. Tobin and on June 28 the crop was already in full bloom. Ascospores were present in the crop throughout the period June 28-July 7 when environmental conditions were conducive to host infection.

There are other factors that might have contributed to the variations in disease levels among tests. Although all trials were within an area of 2200km² centered on Meadow Lake, the amount of precipitation may have been higher at some locations than at others. Soil fertility may also have varied. Plants grown on fertile soil would probably retain

their leaves longer than plants grown on infertile soil. Differences in stand density and lodging may also have been involved. The crops in fields 88-5 and 88-6 were slightly less dense than the crops in the other four fields. More lodging occurred in fields 88-1 and 88-3 than in the other four.

4.3.2.2.3 Field 88-1

Significant differences among rates of benomyl application were detected in this test (Table 4-6). As expected, disease incidence was progressively reduced by increasing the rate of application (Table 4-7). However, significant reductions compared to the check were obtained even by application at 0.15 kg a.i./ha (25% of the recommended rate). The LF3 80 nozzles gave slightly better control than the 11001 nozzles (smaller droplet size) but the difference was not significant at $p=0.05$ (Table 4-7). No significant interaction between nozzle type and rate of application occurred (Table 4-6).

Application of benomyl at late bloom resulted in effective control of stem rot in this test despite the abundance of inoculum at early bloom. However, the efficacy of late bloom application was not demonstrated unequivocally because disease pressure was low.

Table 4-6 Analysis of variance of incidence of stem rot in relation to 2 nozzle types and 4 rates of benomyl application, field 88-1 at Meadow Lake.

Source	df	MS	F
Block	3	14.3	1.17
Nozzle type	1	21.3	1.73
Rate of application	3	335.2	27.35**
Nozzle*rate interaction	3	3.7	0.30
Error	21	12.3	

** Significant at p=0.01.

Table 4-7 The effect of benomyl treatments at late bloom on incidence of stem rot in field 88-1 at Meadow Lake in 1988.

Factor	Treatment	Disease incidence ⁺	Percentage control ^x
Nozzle type	LF3 80	4.2	--
	11001	5.8	--
		SE 0.55	
Rate of application (kg a.i./ha)	check	18.0a [^]	--
	0.15	6.9b	62
	0.30	4.4bc	76
	0.60	3.6c	80
		SE 1.42	

+ Nozzle type means are based on 12 values (check plots were excluded); rate of application means are based on 8 values.

x $\frac{\text{check} - \text{sprayed}}{\text{check}} \times 100$

[^] Values followed by same letter do not differ significantly from one another according to a protected LSD test at p=0.05 after arcsine transformation of the data.

SE = standard error.

4.3.2.2.4 Field 88-3

No significant differences in disease incidence among rates of application were detected in this test (Table 4-8). Application at the recommended rate resulted in a disease reduction of only 33% compared to the check (Table 4-9). The nonsignificant disease control was not surprising because the crop had almost finished flowering at the time the fungicide was applied. At the time of application disease symptoms were evident on leaf blades and petioles, indicating that many infections were already well developed. Inoculum was not abundant until the transition between full and late bloom (Figure 4-2). The fungicide might have been much more effective if applied three days earlier, as originally intended.

Disease incidence was slightly higher in plots sprayed using 11001 nozzles than in plots sprayed using LF3 80 nozzles but the difference was not significant at $p=0.05$ (Table 4-9). There was no significant interaction between nozzle type and rate of application (Table 4-8).

There was significantly more disease in some replicate blocks than in others (Table 4-8). This was probably caused by variations in the amount of lodging. Disease incidence was highest in replicate 4 where the crop was severely lodged and lowest in replicate 1 where the crop was not lodged at all.

Table 4-8 Analysis of variance of incidence of stem rot in relation to 2 nozzle types and 4 rates of benomyl application, field 88-3 at Meadow Lake.

Source	df	MS	F
Block	3	909.0	28.88**
Nozzle type	1	47.3	1.50
Rate of application	3	72.1	2.29
Nozzle*rate interaction	3	4.1	0.13
Error	21	31.5	

** Significant at p=0.01.

Table 4-9 The effect of benomyl treatments at late bloom on incidence of stem rot in field 88-3 at Meadow Lake in 1988.

Factor	Treatment	Disease incidence ⁺	Percentage control ^x
Nozzle type	LF3 80	20.1	--
	11001	23.5	--
		SE 1.89	
Rate of application (kg a.i./ha)	check	28.6	--
	0.15	22.6	21
	0.30	23.4	18
	0.60	19.3	33
		SE 2.34	

+ Nozzle type means are based on 12 values (check plots were excluded); rate of application means are based on 8 values.

x $\frac{\text{check} - \text{sprayed}}{\text{check}} \times 100$

SE = standard error.

4.3.2.2.5 Field 88-4

Disease pressure was moderate in this test. Significant differences among rates of application occurred (Table 4-10). Disease incidence was progressively reduced as the rate of application was increased (Table 4-11). Benomyl gave

Table 4-10 Analysis of variance of incidence of stem rot in relation to 2 nozzle types and 4 rates of benomyl application, field 88-4 at Meadow Lake.

Source	df	MS	F
Block	3	24.0	1.48
Nozzle type	1	81.6	5.02*
Rate of application	3	222.1	13.65**
Nozzle*rate interaction	3	20.3	1.25
Error	21	16.3	

*, ** Significant at $p=0.05$ and 0.01 respectively.

significant control of stem rot at 0.15 kg a.i./ha (25% of the recommended rate). Although significant disease reductions from spraying occurred, a high level of control was not achieved in this test. Even applications at the recommended rate did not reduce disease incidence to half that in the check plots. Inoculum of S. sclerotiorum was present in the crop at early bloom (Figure 4-2). At late bloom disease symptoms were observed on leaf blades and petioles, indicating that many infections were already well developed. Benomyl

Table 4-11 The effect of benomyl treatments at late bloom on incidence of stem rot in field 88-4 at Meadow Lake in 1988.

Factor	Treatment	Disease incidence ⁺	Percentage control ^x
Nozzle type	LF3 80	31.8*	--
	11001	26.2	--
		SE 1.78	
Rate of application (kg a.i./ha)	check	43.6a [^]	--
	0.15	32.8b	25
	0.30	31.1b	29
	0.60	23.0c	47
		SE 2.32	

+ Nozzle type means are based on 12 values (check plots were excluded); rate of application means are based on 8 values.

x $\frac{\text{check} - \text{sprayed}}{\text{check}} \times 100$

* Nozzle type means are significantly differently from one another according to an F test at $p=0.05$ after arcsine transformation of the data.

[^] Values followed by same letter do not differ significantly from one another according to a protected LSD test at $p=0.05$ after arcsine transformation of the data.
SE = standard error.

application at late bloom would not be expected to provide a high level of control under these conditions.

In this test 11001 nozzles were significantly ($p=0.05$) superior to LF3 80 nozzles in reducing disease incidence (Table 4-11). These results differ from those found in fields 88-1 and 88-3 where the difference between nozzle types was not significant. However, in field 88-1 disease pressure was

very low and in field 88-3 significant disease reductions from spraying were not obtained. Thus, there is limited evidence from this study that small spray droplets generated by 11001 nozzles may improve control with benomyl.

Disease index data were also obtained in field 88-4 (Appendix D). Interpretations based on index data were the same as those based on incidence data with two exceptions. With index data, the difference in disease index between 0.15 kg a.i./ha and the check was not significant. The difference between the 2 nozzle types was significant only at $p=0.07$.

4.3.3 Experiment 2

This experiment was similar to that conducted at Melfort in 1987. The disease level in check plots was compared with that in plots sprayed with benomyl at late bloom.

4.3.3.1 Materials and methods

The same experiment was set up in fields 88-5 and 88-(7-10). All five fields were planted to Brassica napus cv. Westar. Each trial was divided into 48 2m x 5m plots of which 24 plots were sprayed at late bloom with benomyl at the recommended rate (0.6 kg a.i./ha) and 24 plots were left untreated. The plots were paired so that a sprayed plot was always adjacent to a check plot. Treatments were applied with a hand-held applicator at 240 kPa and 400 l H₂O/ha using

LF3 80 nozzles. Spray dates were July 5, 7, 11, 16 and 20 for fields 88-10, 88-5, 88-9, 88-8 and 88-7 respectively.

The trials were assessed for disease in August when the plants were nearly mature. Disease incidence was determined by counting the percentage of infected plants in a sample of 200 plants in the center of each plot. In fields 88-(5,9 and 10), disease levels were very low and only the check plots were assessed for disease. In field 88-8, disease severity was also assessed by scoring individual plants for amount of disease using a rating scale (Appendix A). A disease index was calculated for each plot using the formula given in Section 4.2.1.1.2.

4.3.3.2 Results and discussion

The PPI at different growth stages during flowering for all five crops are presented in Figure 4-3. The PPI data are for the sampling sites closest to each fungicide trial. The mean PPI based on all sites in each field were also calculated for each growth stage (Appendix B).

Inoculum was present on the petals at the onset of flowering in fields 88-7 and 88-8 (Figure 4-3). The PPI values were very high (>85) in these two crops at full bloom. In fields 88-5 and 88-9, the PPI values were very low until full and late bloom respectively. The growth stage at which inoculum first appeared in field 88-10 is unknown because the petals were not tested until the crop was in the transition

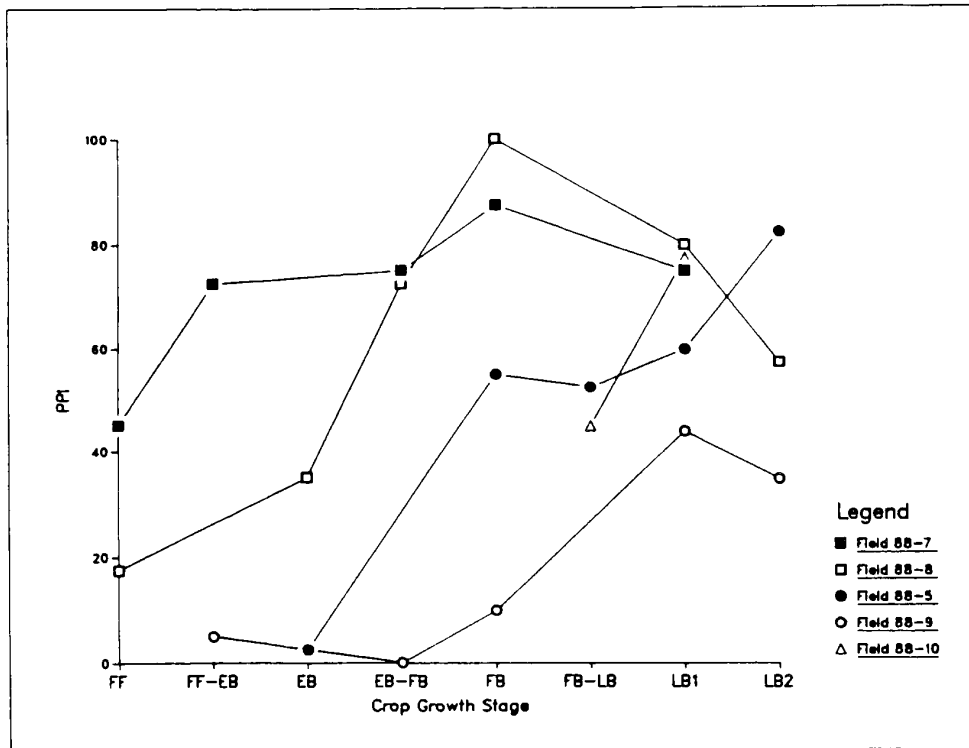


Figure 4-3 Percentage petal infestation (PPI) during the bloom period for five crops, Meadow Lake 1988 (FF = first flower, EB = early bloom, FB = full bloom, LB = late bloom).

between full and late bloom. At this time PPI was already above 40.

The mean disease incidence in the check plots was less than 2 in fields 88-(5,9 and 10) and no useful data were obtained from these tests. In fields 88-7 and 88-8, where inoculum was present from first flower onward, a low level of disease developed. The data from these tests were arcsine transformed and analysed by paired t-tests.

The late bloom application of benomyl significantly reduced disease in both field 88-7 and field 88-8 (Table 4-12). Disease levels in the sprayed plots were approximately half those in the check plots at both locations. Under such low disease pressure, these disease reductions did not represent levels of control that would be acceptable to a farmer. Inoculum had been present in the crops for over 2 weeks before the fungicide was applied. Some of the infections were probably well advanced by late bloom and

Table 4-12 The effect of benomyl application at late bloom on the amount of stem rot in fields 88-7 and 88-8, Meadow Lake 1988.

		Field 88-7	Field 88-8
Disease incidence (SD)	sprayed	8.4 ⁺ (0.9)	7.5 ⁺ (0.8)
	check	17.3 (1.3)	18.5 (2.1)
Percentage control [^]		51	59
Disease index (SD)	sprayed	--	5.1 ⁺ (0.5)
	check	--	11.5 (1.4)
Percentage control [^]			56

All figures represent means of twenty-four replications.

+ Significantly different from check at p=0.01 (paired t-test after arcsine transformation of the data).

[^] $\frac{\text{check} - \text{sprayed}}{\text{check}} \times 100$

SD = standard deviation.

this might have limited control. If more disease had developed late bloom fungicide application would probably have been more effective in fields 88-(5,9 and 10) where inoculum was not detected on the petals until full bloom or later. Unfortunately, the absence of disease in these fields and in fields 88-(2 and 6) discussed in Section 4.3.2, severely limited the conclusions that may be drawn from the 1988 fungicide tests.

5. Effect of Time of Infection on Yield Loss

5.1 Introduction

There is very little information in the literature on the effect of sclerotinia stem rot on yield in rapeseed. Also, in the reports that have been published, the crop growth stage at which infections were initiated was not determined (Krüger and Stoltenberg, 1983; Morrall et al., 1976, 1984a). The purpose of the present work was to compare the effect on yield of infections initiated at early bloom with infections initiated at full or late bloom. If infections initiated late in the bloom period have little effect on yield then fungicide application at late bloom would probably not be economically feasible. A combination of growth chamber and field experiments were conducted to achieve the objective.

5.2 Growth chamber experiment

5.2.1 Materials and methods

5.2.1.1 Planting procedures and environmental conditions

Six seeds per pot of Brassica napus cv. Westar were planted in 15cm clay pots filled with a 3 soil:1 sand:1 peat mixture. Three growth chambers were used and there were 36 pots in each growth chamber. The pots were thinned to 4

seedlings per pot soon after emergence, choosing those seedlings that were well spaced and uniform in size. The pots were thinned to 3 plants per pot and to 2 plants per pot when the plants were in early bud and first flower respectively, choosing those plants that were most similar in development and vigour. The insecticide Piramor 50 (50% a.i.) diluted in water was sprayed to control aphids when the plants were in the early bloom stage.

The plants were grown under controlled environmental conditions. Light intensity was measured with a Li-Cor Model L1-185B photometer when the plants were in the late bloom stage. Fluorescent and incandescent lamps provided a light intensity of approximately $200 \mu\text{E}/\text{m}^2/\text{s}$ at plant height in all three growth chambers. A 16h photoperiod was used. The temperature was maintained at 20C by day and 15C at night except when the plants were flowering, when the temperature was set at 25C by day and 15C at night. The relative humidity (RH) was maintained at 50% by day and 70% at night until the plants were in full bloom when it was raised to 95% (day) and 100% (night) to enhance infection. When the plants were ripening the RH was lowered to 30% (day and night) to hasten maturity. The plants were watered once a day until flowering commenced. At this time the pots were placed in trays containing water and the plants obtained water by capillary rise. The plants were fertilized approximately once a week with 20-20-20 dissolved in water.

5.2.1.2 Inoculation and experimental design

In each pot, one plant was inoculated at either early, full or late bloom and the other plant was not inoculated and served as a covariate. The three treatments (early, full and late bloom) were completely randomized in each growth chamber. Thus, each growth chamber served as a block.

Plants were inoculated using a technique similar to that used successfully by LeCoz (1981) in France. A 5mm mycelial plug was taken from the edge of a 3-4 day old colony of S. sclerotiorum grown on unamended PDA and placed on a single petal in the axil of the second lowest leaf. The leaf axil was wrapped in aluminum foil and moist cotton was pressed against the mycelial plug. Water droplets were added to the cotton twice daily until the lesion had completely girdled the main stem. With this technique, 104 of 108 inoculated plants showed disease symptoms. Symptoms were usually observed on the petioles 2 days after inoculation and on the main stem 3-4 days after inoculation.

In preliminary studies, it was found that plants inoculated with mycelial plugs showed disease symptoms about 5 days earlier than plants inoculated by placing ascospore-infested petals in leaf axils. Because mycelial plugs were 'fast-acting', the early, full and late bloom inoculated plants were inoculated towards the end of their respective growth stages. For example, the late bloom inoculated plants were not inoculated until flowering was almost complete.

Natural infections in the field are caused by ascospores and not by mycelial plugs. Thus, it might have been more desirable to use an inoculation method based on ascospores in this experiment. However, in preliminary studies inoculations with mycelial plugs consistently caused infections but inoculations with ascospore-infested petals did not.

5.2.1.3 Data collection and analysis

When the diseased plants were almost completely ripe the length of lesion on the main stem was measured to the nearest 5mm with a ruler. The pods were picked and placed in small envelopes when the plants were completely mature, air-dried for 1 week and then shelled out to determine seed yield. For each plant the number of pods, seed yield and 100-seed weight were recorded.

The lesion length data were subjected to analysis of variance. The yield data were subjected to analysis of covariance and means were separated using nonorthogonal specific contrast comparisons. For each of the three inoculation treatments the yield of the healthy plants was compared to the yield of the diseased plants using a paired t-test and the percentage reduction from disease was calculated.

5.2.2 Results

The lesion length was greater on full bloom-inoculated plants than on early or late bloom-inoculated plants (Table 5-1) but differences among treatments were not significant at $p=0.05$ (Table 5-2). There was a significant growth stage x growth chamber interaction (Table 5-2). Thus, the differences in lesion length among the three treatments were not consistent among the three growth chambers (Figure 5-1).

The mean seed yield of early bloom-inoculated plants was significantly lower than the mean seed yield of late bloom-inoculated plants according to a specific contrast comparison at $p=0.05$ (Tables 5-3,-4). The mean seed yield of full bloom-inoculated plants was intermediate between the other two treatments but was not significantly different from either of

Table 5-1 Effect of inoculation with S. sclerotiorum at different plant growth stages on lesion length in rapeseed in a growth chamber experiment.

	Inoculation at:		
	early bloom	full bloom	late bloom
Mean lesion length (mm)	265	325	265
Standard error	11.2	11.0	11.2
n	35	34	35

Table 5-2 Analysis of variance of lesion length in relation to 3 periods (growth stages) of inoculation and 3 growth chambers.

Source	df	MS	F
Growth stage (GS)	2	41754	3.56
Growth chamber (GC)	2	20317	4.76*
GS*GC interaction	4	11717	2.75*
Error	95	4267	

* Significant at $p=0.05$.

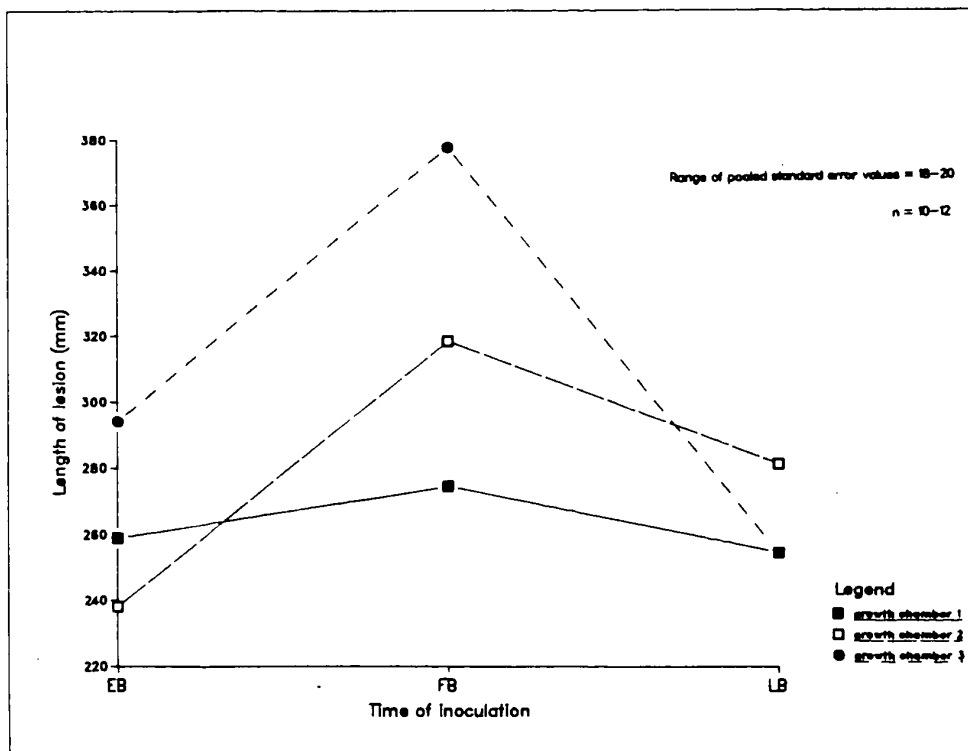


Figure 5-1 Effect of inoculation with *S. sclerotiorum* at different plant growth stages on lesion length in rapeseed in three growth chambers (EB = early bloom, FB = full bloom, LB = late bloom).

Table 5-3 Effect of inoculation with S. sclerotiorum at different growth stages on seed yield of rapeseed in a growth chamber experiment.

	Inoculation at:		
	early bloom	full bloom	late bloom
Seed yield per plant (g) ⁺	0.07	0.28	0.44
Standard error	0.07	0.07	0.07

+ Means are based on 34 values and were adjusted to remove the effect of the covariate (yield of healthy plants).

Table 5-4 Analysis of covariance of seed yield in relation to 3 inoculation periods (growth stages) and 3 growth chambers, and specific contrast comparisons for growth stage effects.

Source	df	MS	F	P
Regression	1	0.6890	22.20	<0.001
Growth stage (GS)	2	1.1544	6.43	0.056
early bloom vs. full bloom	1	0.7128	3.97	0.117
early bloom vs. late bloom	1	2.2974	12.81	0.023
full bloom vs. late bloom	1	0.4458	2.48	0.190
Growth chamber (GC)	2	0.0115	0.37	0.692
GS*GC interaction	4	0.1794	5.77	<0.001
Error	92	0.0311		

them. The overall F test for growth stage was significant only at $p=0.056$ (Table 5-4). Again there was a significant growth stage x growth chamber interaction. The significant

regression indicates that the yield of healthy plants in pots was directly correlated with the yield of diseased plants. Some of the variability in the yield of the diseased plants was due to differences in fertility, moisture conditions etc. among pots.

Analysis of covariance of number of pods and 100-seed weight gave similar results as the seed yield analysis (Appendices E and F). In the 100-seed weight analysis the regression was not significant indicating that this variable was probably less affected by fertility and moisture conditions than number of pods or overall seed yield.

The early, full and late bloom-inoculated plants had significantly lower seed yield, number of pods and 100-seed weight than their healthy counterparts (Table 5-5). The reduction in yield per plant was 93% for the early bloom-inoculated plants. Some of these plants produced no seed at all. The yield reduction was high (58%) even for those plants inoculated at late bloom and was attributed to a decrease in the number of pods as well as a decrease in the weight of the seeds.

5.2.3 Discussion

The lesions on full bloom-inoculated plants were longer than those on early or late bloom-inoculated plants. Many of the early bloom-inoculated plants were stunted by the disease

Table 5-5 Effect of inoculation with S. sclerotiorum at different growth stages on reduction in seed yield, number of pods and 100-seed weight of rapeseed in a growth chamber experiment.

		Inoculated at:	Healthy	Diseased*	Percentage reduction ⁺
<hr/>					
Seed yield in g (SD)	early bloom		1.08 (0.12)	0.07 (0.02)	93
	full bloom		1.15 (0.09)	0.29 (0.03)	75
	late bloom		1.01 (0.09)	0.42 (0.05)	58
Number of pods (SD)	early bloom		16.4 (1.9)	2.4 (0.5)	86
	full bloom		16.3 (1.2)	7.9 (0.6)	52
	late bloom		15.6 (1.3)	10.1 (1.2)	35
100-seed weight in mg (SD)	early bloom		368 (17.4)	143 (10.9)	61
	full bloom		366 (10.1)	186 (7.8)	49
	late bloom		329 (9.1)	223 (10.7)	32

* Difference between means of healthy and diseased plants was significant at $p=0.05$ for all three treatments with all three variables (paired t-tests).

+ $\frac{\text{healthy} - \text{diseased}}{\text{healthy}} \times 100$

SD = standard deviation.

and this may have been a factor involved in the lower lesion length for these plants. The lesions on late bloom-inoculated plants probably did not have time to reach their maximum length before ripening occurred.

The results of this experiment indicate that there is a relationship between yield reduction per plant and the plant growth stage at which infections are initiated. The reduction

in yield per plant was higher when plants were inoculated early as opposed to late in the bloom period. Plants infected at early bloom suffered almost complete yield reduction (93%). Most of this reduction was due to a decrease in the number of pods. The mean number of pods on the early bloom-inoculated plants was 2.4, a reduction of 86% compared to the healthy plants. At the early bloom stage few pods had formed. It appears that once the lesion had completely girdled the main stem, the formation of new pods was prevented or severely curtailed. The yield reduction of the full bloom-inoculated plants (75%) and late bloom-inoculated plants (58%) could be attributed approximately equally to a decrease in the number of pods and a decrease in 100-seed weight. This contrasts with the results of Morrall et al. (1976). They found that percentage reduction in thousand kernel weight of samples from diseased plants closely paralleled percentage yield reduction per plant. Thus, they attributed most of the yield loss to a reduction in seed size.

According to the present results, infections initiated at late bloom can cause considerable yield loss. Because inoculations were made on the main stem the entire inflorescence was always affected by the disease. In the field many late bloom infections probably occur high up on the plant and affect only part of the branching inflorescence. These 'branch infections' would not be as damaging as main stem infections.

5.3 Field experiment

5.3.1 Materials and methods

The experiment was conducted in 1988 using three crops of Brassica napus cv. Westar in the Meadow Lake area. The petal infestation had been monitored at five sites in each field using the procedures described in Section 4.3.1. The three fields chosen for this study differed with respect to the crop growth stage at which substantial inoculum levels were first detected on the petals (Figure 5-2). If a PPI of 30 is arbitrarily considered to be a 'substantial' inoculum level then field 88-4 reached this at early bloom. In fields 88-11 and 88-12, a PPI of 30 was not detected until the crops were in the full and late bloom stages respectively. Since infections cannot occur until inoculum is present, the three crops probably differed in the growth stage at which most infections were initiated. For example, the diseased plant population in field 88-12 probably consisted mainly of late bloom-infected plants since the PPI was low until the late bloom stage.

In each field 25 sites were chosen in August shortly before the crops were swathed. The sites were spaced about 20m apart and were located about 10m from the edge of the field. At each site a search was made for plants with stem rot symptoms. A plant was considered diseased if symptoms

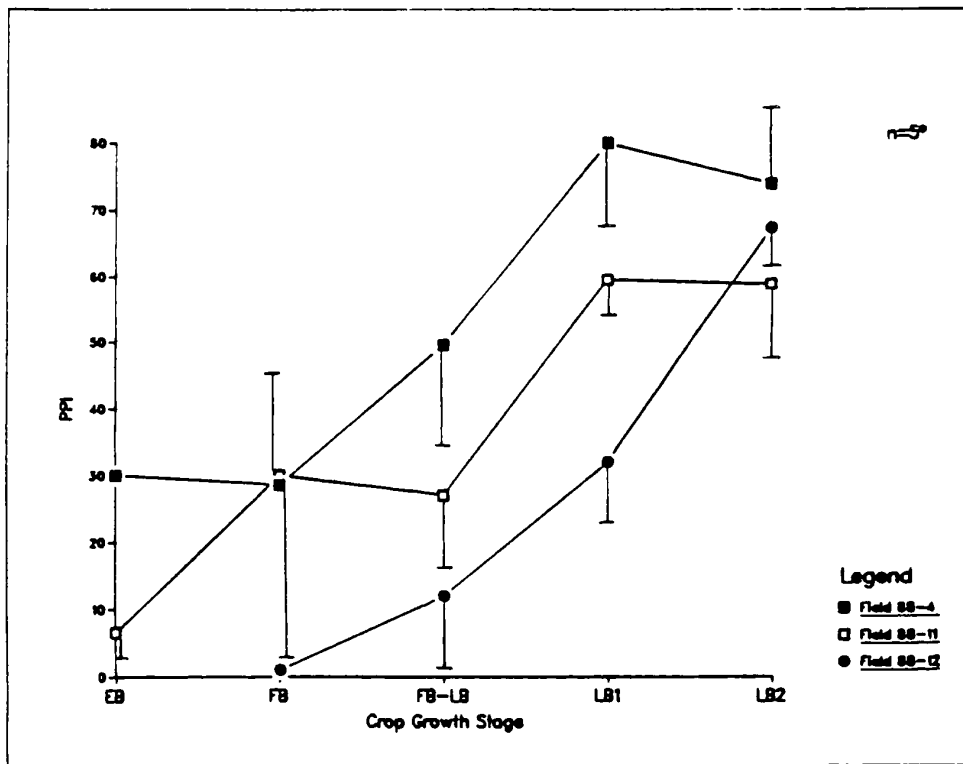


Figure 5-2 Percentage petal infestation (PPI) and standard deviation during the bloom period for the three rapeseed crops in the yield loss experiment, Meadow Lake 1988 (EB = early bloom, FB = full bloom, LB = late bloom).

* All data points represent means of five sites except the early bloom sampling date for field 88-4 where n=1.

were detected on any part of the plant with the exception of the pods. After collecting 100 diseased plants an equal number of healthy plants were pulled from the same area. The diseased and healthy plants were bagged separately. The plants were dried, combined and final seed yields were determined after cleaning and drying the seed. The seed

yields of 100 healthy plants and of 100 diseased plants were thus determined for each site.

The yield data were subjected to analysis of covariance to determine if the yield of diseased plants varied among fields. The covariate was the yield of 100 healthy plants. Non-orthogonal specific contrast comparisons were used to separate means. For each field the yield of the diseased plants and the yield of the healthy plants were compared with paired t-tests and the percentage reduction in yield per plant was calculated.

5.3.2 Results

The mean seed yield of diseased plants was significantly higher in field 88-12 than in the other two fields (Tables 5-6,-7). The significant regression in the analysis of covariance indicates that the yield of the healthy plants was correlated directly with the yield of diseased plants. Thus, some of the variability in the yield of the diseased plants was due to differences in fertility, moisture conditions etc. among sites.

In all three fields the diseased plants were significantly lower yielding than the healthy plants (Table 5-8). The percentage reduction in yield per plant was 37, 32 and 8 for fields 88-4, 88-11 and 88-12 respectively. When data from all fields were averaged, the diseased plants yielded 22% less than the healthy plants.

Table 5-6 Analysis of covariance and specific contrast comparisons of seed yield of rapeseed plants infected with S. sclerotiorum in 3 fields, Meadow Lake 1988.

Source	df	MS	F
Regression	1	101345	47.1*
Field	2	67675	31.4*
88-4 vs. 88-11	1	14	0.01
88-4 vs. 88-12	1	76412	35.8*
88-11 vs. 88-12	1	78563	36.5*
Error	71	2154	

* Significant at $p=0.01$.

Table 5-7 Mean seed yield of diseased rapeseed plants in three fields which differed with respect to the crop growth stage at which inoculum of S. sclerotiorum first became widespread in the canopy, Meadow Lake 1988.

	Field 88-4	Field 88-11	Field 88-12
Inoculum first widespread in crop at:	early bloom	full bloom	late bloom
Seed yield per 100 diseased plants (g)+	182.3	181.1	318.2
Standard error	9.4	12.7	13.5

+ Means are based on 25 values and were adjusted to remove the effect of the covariate (yield of healthy plants).

Table 5-8 Effect of stem rot on yield of rapeseed plants in three fields which differed with respect to the crop growth stage at which inoculum of S. sclerotiorum first became widespread in the canopy, Meadow Lake 1988.

	Inoculum first widespread in crop at:	Seed yield per 100 plants in g (SD)		Percentage reduction ⁺
		healthy	diseased*	
Field 88-4	early bloom	276 (13)	174 (13)	37
Field 88-11	full bloom	179 (11)	122 (6)	32
Field 88-12	late bloom	418 (18)	386 (15)	8

Figures in the table are the means of 25 replicates.

* Difference between means of healthy and diseased plants was significant at $p=0.05$ for all three fields.

+ $\frac{\text{healthy} - \text{diseased}}{\text{healthy}} \times 100$

SD = standard deviation.

5.3.3 Discussion

The effect of disease on yield was very slight in field 88-12 compared to fields 88-4 and 88-11. The appearance of inoculum was delayed until almost late bloom in field 88-12. In fields 88-4 and 88-11 at least some inoculum was present on the petals from the early bloom stage onward. Therefore, the results indicate that infections initiated at the late bloom stage are less damaging than infections initiated earlier in the bloom period. The yield reduction per plant in field 88-4 (37%) was slightly higher than in field 88-11 (32%). Field 88-4 had a higher PPI at early bloom than field

88-11. The diseased plant population of field 88-4 probably consisted of relatively more early and full bloom-inoculated plants than did the diseased plant population of field 88-11.

The extremely low yield reduction per plant (8%) for field 88-12 does not support the results of the growth chamber experiment. The plants inoculated at late bloom in the growth chamber yielded 58% less than the healthy plants. There are a number of possible factors that might have caused the discrepancy. As mentioned earlier, infections in the growth chamber were always on the main stem but 'branch infections' were common in the field. Some infections in the field may have been initiated after flowering was complete, although inoculum was present earlier. These 'extremely late' infections would probably not be damaging. Finally, environmental conditions in the growth chamber probably favored disease development more than conditions in the field.

Morrall et al. (1984a) studied the relationship between yield of rapeseed and stem rot incidence using data from fungicide trials and commercial fields collected over several years. They found that the reduction in yield per plant from the disease was generally 40-50%. In the above study inoculum was present in all the fields at early bloom (R.A.A. Morrall, personal communication). In the present study the yield reduction per plant was lower than 40% in all three fields. Even in field 88-4, where the PPI was 30 at early bloom, the yield reduction per plant was only 37%. In another

field in the Meadow Lake area in 1988 (field 88-13) an observation was made independently by several co-workers that might aid in the interpretation of results. It appeared that smaller plants were more likely to escape infection than larger, highly branched plants. If this had happened to any extent in the three fields selected for yield loss assessment, the yield reduction from disease would have been underestimated.

The results of the present study were somewhat comparable to those of Morrall et al. (1976). They separated diseased from healthy plants in four fields after the crops were swathed and found that the reduction in yield per plant varied from 22.2% to 54.5%. However, this method of yield loss assessment was similar to that used in the present study and might also have underestimated losses.

6. Incidence-Severity Relationships

6.1 Introduction

Plant pathologists usually use either disease incidence or disease severity measurements in disease assessment. Seem (1984) defined disease incidence as "the proportion (0 to 1) or percentage (0 to 100) of diseased entities within a sampling unit" and severity as "the quantity of disease affecting entities within a sampling unit". Disease severity measurements usually have a closer relationship with yield loss than incidence measurements because they take into account the relative area of plant tissue affected by the disease. However, disease incidence is easier to measure than severity and can also be more precisely determined. In recent years, incidence-severity (I-S) relationships have been studied with many different pathosystems (Seem, 1984). The main application has been simplification of disease assessment since a quantifiable, consistent I-S relationship allows researchers to estimate severity based on incidence measurements.

The I-S relationship for sclerotinia stem rot of rapeseed has seldom been studied. It was investigated in the present study to determine if assessment of stem rot could legitimately be simplified. In several of the field experiments in 1987 and 1988 both disease incidence and

disease severity were measured in each plot. Disease incidence was simply the percentage of infected plants. The disease severity measure was the disease index calculated after scoring individual plants for amount of disease using a rating scale (Appendix A). Regression analysis was used to study the relationship of severity on incidence.

6.2 Materials and methods

Incidence and index data collected from plots at eight locations were used to determine an overall I-S relationship (Table 6-1). The overall data set was classified according to location, year, fungicide treatment (check vs. early bloom spray vs. late bloom spray) and crop growth stage at which inoculum was first present (early vs. full or late bloom) and separate regressions were calculated for each subset. A test of the homogeneity of slope coefficients was used to compare I-S relationships (Gomez and Gomez, 1984).

6.3 Results

The R^2 value for the overall data set was 95.5%, indicating a strong relationship between disease incidence and disease severity. The regression equation disease severity = $0 + 0.692 \times$ disease incidence described the relationship. The slopes for regression equations were not significantly different between years (Table 6-2). Thus, a

Table 6-1 Data used to study incidence-severity relationship for stem rot in rapeseed; classification according to location, year, fungicide treatment and initial inoculum arrival.

Location	Year	Fungicide treatment ⁺	Initial inoculum arrival [^]	n
Canwood	1987	1,3	1,2	51
Field 87-1	1987	1,3	2	24
Field 87-2	1987	1,3	2	24
Field 87-3	1987	1,2,3	1	80*
Field 88-1	1988	1	1	8
Field 88-3	1988	1	2	8
Field 88-4	1988	1,3	1	32
Field 88-8	1988	1,3	1	48

+ 1=check, 2=treatment at early bloom, 3=treatment at late bloom.

[^] 1=inoculum present from early bloom onward, 2=appearance of inoculum delayed until full or late bloom.

* Includes data from an adjacent fungicide trial by Morrall and Verma (1987).

n = number of plots.

plot of disease severity against disease incidence using 1988 data illustrates the I-S relationship (Figure 6-1).

The slopes from the regression equations were not significantly different among the three fungicide treatments (Table 6-2). The growth stage at which inoculum was first present in the crop also did not have a significant effect. Significant differences in slope coefficients were detected among fields. The slope coefficient for field 88-3 (0.843) was relatively high compared to the other seven fields.

Table 6-2 Parameters from regression analyses of disease severity (DS) on disease incidence (DI) for stem rot in rapeseed according to fungicide treatment, initial inoculum arrival, location and year.

		Range of DI values	Range of DS values	R ² (%)	Slope	n
Fungicide treatment	check	2.0-66.0	1.4-51.0	92.6	0.694	116
	EB spray	0-37.1	0-30.0	91.0	0.715	44
	LB spray	0-49.0	0-38.5	98.6	0.707	115
Initial inoculum arrival	EB	0-66.0	0-51.0	94.6	0.687	178
	FB or LB	0-47.5	0-39.8	96.5	0.689	97
Location	Canwood	0-39.0	0-25.4	98.9	0.651*	51
	Field 87-1	0.5-41.5	0.1-27.6	99.2	0.638	24
	Field 87-2	1.0-36.5	0.3-24.1	97.8	0.598	24
	Field 87-3	0-66.0	0-51.0	93.0	0.694	80
	Field 88-1	11.0-31.5	7.0-20.9	98.8	0.683	8
	Field 88-3	5.5-47.5	4.0-39.8	99.1	0.843	8
	Field 88-4	11.5-53.0	8.1-37.9	91.5	0.629	32
	Field 88-8	2.0-43.0	1.1-28.9	98.4	0.647	48
Year	1987	0-66.0	0-51.0	94.7	0.687	179
	1988	2.0-53.0	1.1-39.8	96.6	0.708	96

* Significant differences in slopes, $p=0.05$, were detected among locations based on a coefficient comparison test.

n = number of observations.

EB = early bloom, FB = full bloom, LB = late bloom.

Unfortunately, the regression equation for field 88-3 was based on only eight data points. Interestingly, field 88-3 was the only field of eight that was planted to Brassica campestris.

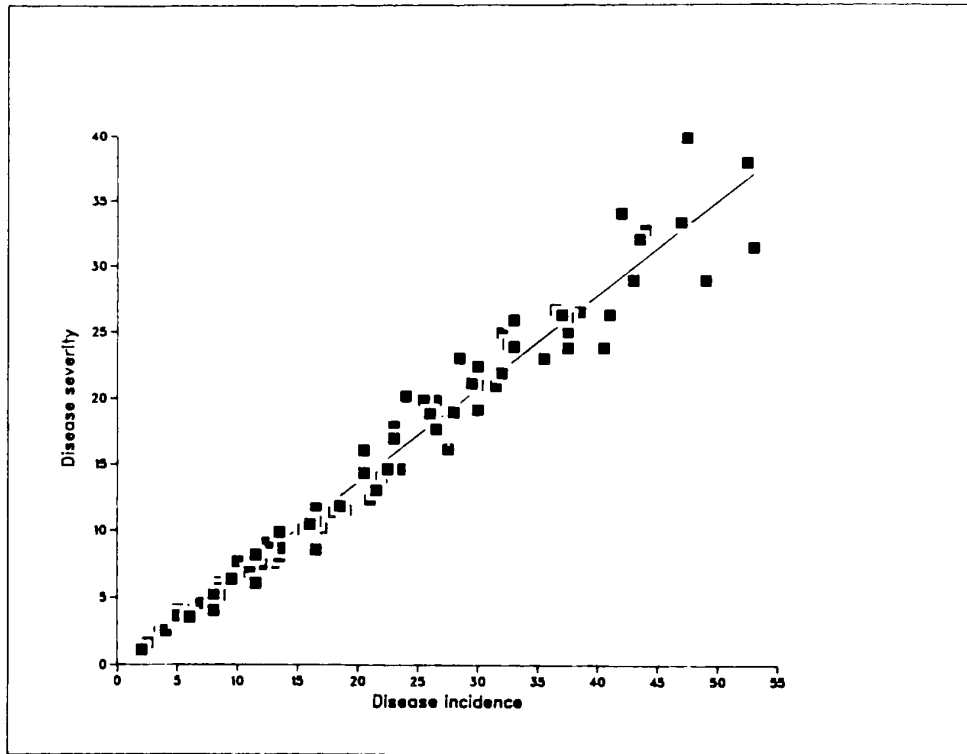


Figure 6-1 Regression of disease severity (% disease index) vs. % disease incidence for stem rot in rapeseed in 1988.

6.4 Discussion

The results indicate that there is a strong, consistent relationship between disease incidence and disease severity for stem rot in rapeseed. Transformation of data was not required to linearize the relationship. The R^2 values were higher than 97.5% at all locations except fields 87-3 (93.0%) and 88-4 (91.5%). In those two fields disease ratings were performed by several people and inconsistencies among individuals may have contributed to the slightly lower R^2 values.

A factor that theoretically could affect the I-S relationship is the crop growth stage at which infections are initiated. A crop in which inoculum was present at early bloom would be expected to include more severely diseased plants (ie. categories 3 and 4, Appendix A) than a crop where the appearance of inoculum was delayed until full or late bloom. With early bloom infection, the fungus has more time to spread systemically within the plant and cause premature ripening and/or seed shrivelling. In addition, the entire inflorescence is generally affected by the disease since infections usually occur lower on the plant because lower leaves are still attached. The slope coefficient, which reflects the rate of increase of severity with increasing incidence would be expected to be higher for crops infected at early compared to full or late bloom. However, the results of the present study indicate that this may not be the case. The slope coefficient for plots where inoculum was present at early bloom (0.687) was actually slightly lower than that for plots where the appearance of inoculum was delayed until at least full bloom (0.689) (Table 6-2).

The slope coefficients were not significantly different between years, and with the exception of field 88-3, the slope coefficients for the different fields were very similar (Table 6-2). Thus, it appears that factors such as rainfall, stand density and inoculum level probably have little effect on the I-S relationship. Fungicide application also does not appear

to affect the relationship since no significant differences in slope coefficients were detected among the three fungicide treatments (Table 6-2). There is limited evidence from this study to suggest that the I-S relationship for Brassica napus cultivars may be different from that for B. napus cultivars.

Collection of only disease incidence data reduces labor requirements considerably in large scale field experiments. It also allows the use of relatively unskilled labor because disease assessment only involves differentiating the presence or absence of disease. According to the results of the present study, researchers working with Brassica napus cv. Westar could omit performing disease ratings without a loss of information. If there was a need in practice to determine disease severity, these values could be predicted using the equation $\text{disease severity} = 0 + 0.692 \times \text{disease incidence}$. The data collected in this study were mainly concentrated on low levels of disease. In future work in this area a wider range of data should be collected. More research is also required to clarify cultivar effects on the I-S relationship.

7. General Discussion

7.1 Feasibility of fungicide application at late bloom

At present, the application of foliar fungicides is considered to be the best method for controlling stem rot of rapeseed in western Canada (Morrall et al., 1985; Thomson et al., 1984). According to current recommendations, spraying should be conducted at either early or full bloom (Thomas, 1984). When moisture conditions are poor during the bud and first flower stages, applying a fungicide at early or full bloom might not seem to be justified since crops would be forecast to be at low disease risk. However, if moisture conditions improve as flowering progresses moderate or high disease levels may still result (Turkington, 1988). The main objective of the present study was to determine if fungicide application at late bloom can effectively control the disease in this situation.

The efficacy of benomyl application at late bloom was evaluated in field experiments. Disease levels were high enough to warrant collection and analysis of disease incidence data at 4 of 5 locations in 1987 and 5 of 10 locations in 1988 (Table 7-1). Application of benomyl at the recommended rate (0.6 kg a.i./ha) significantly reduced disease incidence compared to the check at all locations except in field 88-3 at Meadow Lake. Excellent control was achieved in four tests.

Table 7-1 Synoptic table of results of field experiments evaluating the efficacy of fungicide application at late bloom in 1987 and 1988.

Experiment	Approximate growth stage when inoculum was first widespread in crop ⁺	Rate of benomyl applied (kg a.i./ha)	% control* in relation to relevant check
Canwood	no inoculation	0.30	76
		0.60	85
	early bloom	0.30	63
		0.60	71
	full bloom	0.30	77
		0.60	88
	late bloom	0.30	83
		0.60	93
Field 87-1	late bloom	0.60	90
Field 87-2	full bloom	0.60	83
Field 87-3	late bloom	0.60	39
Field 88-1	early bloom	0.15	62
		0.30	76
		0.60	80
Field 88-3	late bloom	0.15	21
		0.30	18
		0.60	33
Field 88-4	early bloom	0.15	25
		0.30	29
		0.60	47
Field 88-7	early bloom	0.60	51
Field 88-8	early bloom	0.60	59

+ See Section 4 for experimental details.

* $\frac{\text{disease incidence (DI) in check} - \text{DI in treatment}}{\text{DI in check}} \times 100$

In fields 87-(1 and 2) at Melfort, field 88-1 at Meadow Lake and in the Canwood experiment, application at the recommended rate reduced disease incidence by $\geq 80\%$ compared to the check.

There is limited evidence from the experiments in 1987 to suggest that late bloom application is more effective when ascospores are not present in the crop until at least full bloom. At Melfort, the percentage control was only 39 in field 87-3 where ascospores were widespread in the crop from early bloom onward. This level of control was much lower than that achieved in field 87-2 (83%) and field 87-1 (90%) where inoculum was not present until full and late bloom respectively. These results were not supported by those from the Canwood experiment. At Canwood plants were artificially inoculated at either early, full or late bloom. Effective control was achieved in plots inoculated at early bloom as well as in plots inoculated at full or late bloom. However, many infections in early bloom-inoculated plots may not have been initiated until approximately late bloom (Section 7.2) and this would have affected the results.

Unfortunately, very little information on the effect of timing of inoculum arrival on the efficacy of late bloom fungicide application was obtained from the 1988 field tests. With the exception of the test in field 88-3, disease developed only where inoculum was present from early bloom onward. The percentage control using the recommended rate of benomyl was 80, 47, 51 and 59 for fields 88-(1,4,7 and 8)

respectively. These disease reductions were higher than those obtained in field 88-3 (33%) where a high level of inoculum was not detected until almost late bloom. However, in field 88-3 the crop had almost finished flowering by the time the fungicide was applied and this probably contributed to the poor control.

The results of the present study support Krüger's (1973) statement that spraying should coincide with ascospore release for maximum stem rot control. When inoculum is abundant in a crop at early bloom, many infections will be well advanced by late bloom and this will limit the efficacy of late bloom application. In this situation the best time to spray is at early or full bloom before the fungus can become established in the plant.

The rate of application of benomyl was varied in several tests to determine if a high level of control could be achieved by spraying with less than the recommended rate. At Canwood and in fields 88-1 and 88-4 at Meadow Lake, application at 0.3 kg a.i./ha (half-rate) significantly reduced disease incidence compared to the check. In the Canwood and field 88-1 tests there was no significant difference in control between half- and recommended rate applications although in both cases disease incidence was slightly higher in plots sprayed with the half-rate. In field 88-4, applications at 0.3 kg a.i./ha were considerably less effective than those at the recommended rate. This may have

been a result of the higher disease pressure in this field. Applications at 0.15 kg a.i./ha (quarter-rate) were tested in fields 88-(1,3 and 4) and significant disease reductions compared to the check were obtained in fields 88-1 and 88-4. The above results support those of Morrall (1988), Morrall and Verma (1987, 1988) and Morrall et al. (1984b). Application of benomyl at reduced rates provided a high level of stem rot control under low disease pressure.

In field 88-4, treatments applied with 11001 nozzles gave significantly better disease control than those applied with LF3 80 nozzles. The spray droplets generated by 11001 nozzles are smaller than those produced by LF3 80 nozzles. Morrall and Verma (1987) and Morrall (1988) also found that excellent control of stem rot could be achieved using nozzles generating small spray droplets. Rogers et al. (1988) found that small spray droplets have a higher deposit efficiency on rapeseed than larger droplets. Therefore, small spray droplets may improve control with benomyl provided application is made under relatively calm conditions to avoid wind drift.

Fungicide application at late bloom should be considered only in fields where a high level of inoculum is present at full or late bloom, but not earlier. In order to justify chemical control, the gains in yield from control should at least cover the costs of spraying. Therefore, late bloom application would probably never be economical unless infections initiated at full bloom or later have a

considerable effect on yield. The results of the growth chamber experiment indicate that 'late infections' can be severely damaging. The yield reduction per plant from the disease was 58% for plants inoculated at late bloom. However, in the field experiment the yield reduction per plant was less than 40% in all 3 fields. In field 88-12, where a high PPI was not detected until late bloom, the yield reduction per plant was only 8%. Because of the discrepancy in results between the growth chamber and field experiment, the effect on yield of infections initiated late in the bloom period is still unclear.

There are also other factors that must be considered when determining the economic feasibility of late bloom application. Morrall et al. (1983) estimated that the loss from wheel damage was 4% when spraying was done at full bloom with a tractor-mounted sprayer. This loss would probably be higher if application were made at late bloom rather than at an earlier growth stage. The use of a commercial 'spra-coupe' or other high clearance type of equipment, such as is widely available in Europe, might help minimize this type of loss.

Stem rot can cause quality as well as yield losses (Dueck and Sedun, 1983; Thomson and Stelfox, 1983). Therefore, an indirect benefit of fungicide application is reduced dockage resulting from fewer shrivelled seeds and fewer sclerotia harvested with the seed. This is especially relevant to seed

growers. They would probably be more willing than other growers to undertake chemical control at late bloom.

The present study has shown that effective stem rot control can be achieved by spraying at late bloom. However, despite its proven efficacy more research is required in a few areas (Section 7.3) before it can be considered an economic option for rapeseed growers.

7.2 Limitations of experimental results

The value of agricultural field experiments is often highly weather-dependent. This is especially true in studies with Sclerotinia sclerotiorum, a pathogen requiring prolonged moist conditions for infection. In the present study, dry conditions during flowering resulted in the absence of disease at Saskatoon in 1987 and extremely low levels of disease in fields 88-(2,5,6,9 and 10) in 1988. No useful data were obtained from any of these tests. The low disease levels in the five fields in 1988 were especially unfortunate because inoculum of S. sclerotiorum was not abundant in these fields until at least full bloom. It is in this situation that fungicide application at late bloom is most likely to be effective. All of the 'successful' fungicide tests in 1988, with the exception of that in field 88-3, were conducted in fields where inoculum levels were fairly high at early bloom. Ironically, in field 88-3 spraying was delayed until the crop had almost finished flowering because of high winds and heavy

rain. This undoubtedly contributed to the low levels of control achieved in this test. If the weather had been moister during mid- and late-July (Appendix C), disease might have developed in all tests and a more thorough evaluation of fungicide application at late bloom could have been made.

Although significant reductions in disease were achieved with fungicide application at late bloom in some tests, corresponding yield increases from spraying were not demonstrated. However, this was not considered to be a serious limitation because results from the present study (Section 5) and other studies (Krüger and Stoltenberg, 1983; Morrall et al., 1976, 1984a) have shown that stem rot reduces yield in rapeseed. Yields were measured in the tests in fields 87-2 and 87-3 at Melfort in 1987 but the differences between check and sprayed plots were not significant (Table 4-5). Yield data were not collected in the other fungicide tests because disease levels were quite low and significant differences in yield among treatments would have been difficult to demonstrate.

The experiment dependent on artificial inoculation conducted at Canwood in 1987 (Section 4.2.1) also had limitations. Ascospores of S. sclerotiorum are capable of surviving up to 12 days in the field at low RH (Caesar and Pearson, 1983; Grogan and Abawi, 1975). Although ascospores were applied to some plots at early or full bloom, infections may not have been initiated in these plots until late bloom

when environmental conditions were more favorable. Many infections at Canwood were caused by 'background inoculum' and not by the artificially applied inoculum. The 'background inoculum' probably did not arrive in the crop until approximately late bloom because soil conditions were dry and germination of sclerotia would have been prevented until the crop was almost in full bloom. Therefore, the diseased plant populations in early and full bloom-inoculated plots may have mainly consisted of plants that were infected at late bloom instead of at early or full bloom, as intended. As a consequence, no firm conclusions could be drawn about effects of the main factor, ie. period of inoculation.

In retrospect, successful inoculation of plants at early, full or late bloom, as was attempted at Saskatoon and Canwood in 1987 is extremely difficult. It would require the maintenance of moist conditions in the canopy for a period of sufficient length to ensure infection at each of the three periods of inoculation. This would be an unusual occurrence in Saskatchewan, a semi-arid region, unless irrigation water were used to supplement natural precipitation. In the Saskatoon test, the plants were frequently moistened (usually at least once a day) throughout the bloom period using a sprinkler irrigation system. However, since no disease developed it is probable that either irrigation was not frequent enough or the water droplets washed inoculum off petals, thereby reducing the likelihood of infection. The

chance of success would probably have been greater if the irrigation water had been applied as a mist and at regular intervals using a timer. This type of system has been used recently with moderate success to infect rapeseed with S. sclerotiorum at Saskatoon (D.L. McKenzie, personal communication).

Underestimation of losses was a possible limitation of the field experiment in which the effect of time of infection on yield loss was evaluated. Larger and highly branched plants probably had a greater potential to become infected than smaller plants simply because there were more infection sites (eg. leaves, leaf axils). Since large plants are normally higher yielding than small plants, the yield of the diseased plants at each site may have been slightly higher than if plants of all sizes had been equally infected. This would have reduced the difference between the yields of healthy and diseased plants and would have caused an underestimation of yield loss. This problem might have been avoided if, at each site, the healthy plants had been selectively chosen to match the diseased plants in size. However, this procedure would have been very time-consuming and difficult to achieve without bias.

The main limitation of the growth chamber experiment was that the conditions were somewhat artificial. Plants were infected by mycelium of S. sclerotiorum and not by ascospores. Furthermore, environmental conditions in the growth chamber

did not perfectly simulate field conditions. For example, the RH was maintained at 95% by day and 100% at night for most of flowering to enhance infection. In the field the RH would never be as high for such a long period of time. Because of these limitations, caution must be exercised when applying results from the growth chamber to the field.

7.3 Suggested future work

More field experiments need to be conducted to clarify the effect on yield of infections initiated at early, full and late bloom. The procedures need to be changed slightly from those used in the present study to avoid underestimation of yield loss. The relationship between PPI at different flowering stages and final disease incidence is currently being investigated by T.K. Turkington (unpublished) in order to understand better the relative importance of high inoculum levels at full or late bloom as opposed to early bloom. The results of these studies will provide growers with an indication of the disease risk of a crop in which the appearance of a high inoculum level is delayed until at least full bloom. This information is essential for a complete evaluation of fungicide application at late bloom.

Further investigation into reduced rates of application is needed. When a high PPI is not present until full or late bloom, high disease incidence might be a rare occurrence and spraying at half- or even quarter- the recommended rate may

result in adequate control. Nevertheless, the effectiveness of reduced rate applications under high disease pressure should also be determined. The question of the 'best' nozzle type needs to be resolved. Finally, the efficacy of late bloom application might also be investigated for other fungicides, including iprodione.

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Appendix A.

Disease rating system used in field experiments.

Numerical value	Description
0	No disease
1	Non-girdling lesion(s) on stem, or lesion(s) on leaf, pod, or pedicel
2	Stem girdled, no premature ripening
3	Plant prematurely ripe, plump seed
4	Plant prematurely ripe, shrivelled seed

Appendix B.

Percentage petal infestation (PPI) during the bloom period and spray date for ten crops, Meadow Lake 1988.

Field	Sampling date	Growth stage ⁺	Mean PPI* (SD)	Spray date
88-1	July 4	EB	44.0 (11.0)	July 18
	7	EB-FB	72.5 (5.3)	
	10	FB	68.9 (11.6)	
	14	FB	71.0 (8.4)	
	16	FB-LB	88.5 (3.4)	
	20	LB	45.0 (23.3)	
88-2	June 29	EB	1.5 (2.2)	July 9
	July 1	EB-FB	18.0 (4.1)	
	4	FB	36.2 (10.4)	
	7	FB-LB	69.0 (7.8)	
	11	LB	95.5 (2.7)	
	15	LB	89.7 (6.5)	
88-3	June 28	FB	8.3 (9.5)	July 7
	30	FB-LB	42.9 (20.7)	
	July 2	LB	46.0 (9.1)	
	6	LB	80.2 (6.6)	
88-4	June 28	EB	30.0 n=1	July 9
	July 2	FB	28.5 (25.0)	
	6	FB-LB	49.5 (14.7)	
	10	LB	80.0 (11.5)	
	14	LB	73.9 (11.8)	
88-5	June 28	EB	3.8 (2.5)	July 7
	July 2	FB	23.0 (19.6)	
	6	FB-LB	26.5 (15.3)	
	10	LB	50.0 (7.9)	
	14	LB	52.0 (19.8)	
88-6	June 28	EB	5.0 n=1	July 8
	July 1	FB	19.9 (5.6)	
	4	FB	32.5 (10.0)	
	7	FB-LB	71.9 (11.9)	
	11	LB	71.5 (10.2)	
	15	LB	53.0 (5.2)	

Appendix B (cont'd).

Field	Sampling date	Growth stage ⁺	Mean PPI [*] (SD)	Spray date
88-7	July 4	FF	21.0 (15.5)	July 20
	7	FF-EB	51.7 (19.4)	
	11	EB-FB	78.9 (6.3)	
	15	FB	73.4 (16.9)	
	19	LB	69.5 (5.7)	
88-8	July 1	FF	8.5 (5.8)	July 16
	4	EB	18.2 (12.6)	
	7	EB-FB	67.7 (6.1)	
	11	FB	91.0 (7.2)	
	16	LB	81.4 (9.4)	
	19	LB	56.1 (14.6)	
88-9	June 28	FF-EB	3.5 (2.2)	July 11
	30	EB-FB	0 (0)	
	July 3	FB	13.5 (7.4)	
	6	LB	43.3 (14.0)	
	10	LB	51.0 (9.1)	
88-10	July 1	FB-LB	45.0 n=1	July 5
	3	LB	80.0 n=1	

+ FF = first flower, EB = early bloom, FB = full bloom and LB = late bloom.

* Means are based on samples from five sites in each field unless indicated differently.

SD = standard deviation.

Appendix C.

Rainfall and maximum daily temperature data obtained from Atmospheric Environment Service, Environment Canada for June and July, Meadow Lake 1988.

Date	Rainfall (mm)	Max. temp. (C)	Date	Rainfall (mm)	Max. temp. (C)
June 1	0	23	July 1	0	23
2	1.2	22	2	4.4	23
3	0.6	32	3	5.4	21
4	2.6	29	4	0	21
5	0.6	29	5	26.4	23
6	1.2	25	6	2.6	22
7	0	18	7	0.2	19
8	0.6	18	8	0	18
9	2.6	22	9	0	14
10	3.0	28	10	0	19
11	0	21	11	0	24
12	0.4	14	12	4.4	20
13	0	18	13	4.6	20
14	3.6	16	14	22.2	25
15	0	24	15	0	22
16	0	27	16	0	21
17	0	31	17	0	22
18	0	26	18	2.6	21
19	0	22	19	0	22
20	0	25	20	0	21
21	0	24	21	0	22
22	0	22	22	2.6	26
23	0.2	26	23	0	23
24	5.6	21	24	0	24
25	0	27	25	0.6	24
26	0	26	26	0	30
27	0	28	27	0.4	25
28	0.4	25	28	0	24
29	41.4	19	29	0	24
30	1.8	22	30	4.8	23
			31	0	23

Total June rainfall = 65.8mm Total July rainfall = 81.2mm

Appendix D.

The effect of benomyl treatments at late bloom on disease index of stem rot in field 88-4 at Meadow Lake in 1988.

Factor	Treatment	Disease index ⁺	Percentage control ^x
Nozzle type	LF3 80	22.4	--
	11001	18.9	--
		SE 1.34	
Rate of application (kg a.i./ha)	check	28.4a [*]	--
	0.15	23.9ab	16
	0.30	21.6b	24
	0.60	16.5c	42
		SE 1.74	

+ Nozzle type means are based on 12 values (check plots were excluded); rate of application means are based on 8 values.

x $\frac{\text{check} - \text{sprayed}}{\text{check}} \times 100$

* Values followed by same letter do not differ significantly from one another according to a protected LSD test at $p=0.05$ after arcsine transformation of the data.
SE = standard error.

Appendix E.

Analysis of covariance of number of pods in relation to 3 inoculation periods (growth stages) and 3 growth chambers, and specific contrast comparisons for growth stage effects.

Source	df	MS	F	P
Regression	1	288.0	18.87	<0.001
Growth stage (GS)	2	581.0	5.14	0.079
early bloom vs. full bloom	1	516.4	4.57	0.099
early bloom vs. late bloom	1	1112.5	9.84	0.035
full bloom vs. late bloom	1	112.9	1.00	0.374
Growth chamber (GC)	2	27.6	1.80	0.170
GS*GC interaction	4	113.1	7.41	<0.001
Error	92	15.3		

Appendix F.

Analysis of covariance of 100-seed weight in relation to 3 inoculation periods (growth stages) and 3 growth chambers, and specific contrast comparisons for growth stage effects.

Source	df	MS	F	P
Regression	1	4167	1.71	0.195
Growth stage (GS)	2	37279	5.82	0.066
early bloom vs. full bloom	1	22033	3.44	0.137
early bloom vs. late bloom	1	72686	11.34	0.028
full bloom vs. late bloom	1	26002	4.06	0.114
Growth chamber (GC)	2	8748	3.58	0.032
GS*GC interaction	4	6409	2.62	0.041
Error	78	2442		