Clubroot of vegetable brassicas: a review

A report prepared for
The Fresh Vegetable Research & Development Committee, Vegetable & Potato Growers' Federation Inc.

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December 1994

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Frontispiece: Brassica plant heavily infected with clubroot (*Plasmodiophora brassicae*)
Control of clubroot of vegetable brassicas has been identified as an important priority for New Zealand fresh vegetable growers. Recent scientific literature on the disease has been reviewed, with particular reference to advances in control of clubroot. The disease has been difficult to control adequately because the causative organism (*Plasmodiophora brassicae*) has a wide host range, can proliferate rapidly, can survive for long periods (several years) in soil, and is difficult to control with pesticides. The best prospects for control of the disease are to use available knowledge in an integrated strategy involving cultural, chemical and disease resistance methods. Future research should concentrate on the assessment and use of resistance to the disease in vegetable brassica cultivars, investigation of recently available chemicals that show promise for control of clubroot and similar diseases, and possibly the prospects for biological control.
2 INTRODUCTION

Clubroot is a very important disease which can cause severe damage to vegetable and forage brassica crops. The disease was originally thought to occur only in the temperate areas of the world (Karling 1968), but has now been recorded in all regions where brassica crops are grown. A recent survey of fresh vegetable growers by the Fresh Vegetable Sector of the New Zealand Vegetable & Potato Growers' Federation Inc. (VegFed) identified control of clubroot of brassicas as a major priority for the fresh vegetable industry in New Zealand.

A review of current knowledge on clubroot of brassicas has been undertaken. The review was commissioned by the Fresh Vegetable Research & Development Committee of VegFed to provide growers with up-to-date information on the disease, and as the first step in a research project to develop better control strategies for the disease than are currently being used by New Zealand vegetable brassica growers.

The review was undertaken using standard methods of electronic and manual searching of published scientific and popular literature on clubroot of vegetable brassicas, emphasising papers and reports published during the last decade. An exhaustive review of early literature on the disease published by Karling (1968) remains the definitive collection of knowledge on clubroot.

The present report summarises relevant information on clubroot, and includes a brief outline of the disease, the causative organism, and methods that have been investigated for clubroot control. The report will be used as the basis for publications for growers to provide them with current information on clubroot control strategies.
3 THE DISEASE

3.1 History

Clubroot has long been recognised as a very important disease of brassicas used as vegetables for human consumption and as fodder for domestic animals. Historical records show that the disease was known in Italy in the 4th Century AD (Crisp et al. 1989), and was spread through the rest of Europe, possibly on turnips. Clubroot was recorded in Spain in the 15th Century (Karling 1968), in southern Germany in the 16th Century (Crisp et al. 1989), and was first reported in Britain in 1750, in France in 1820, in North America in 1852 (Karling 1968), and in Japan in 1892 (Yoshikawa 1983). In New Zealand, the disease was first recorded on cabbage, cauliflower and forage brassicas in 1894, and probably was widespread in this country at that time (Kirk 1894).

3.2 Economic importance

Clubroot (also known as finger and toe, knolvoet, hernie du chou, kohlhernie, klomprot, and kampoustrnaja kila; Buczacki 1979) can cause severe economic losses in vegetable brassica production. It can reduce marketable yields from, or sometimes completely destroys, cabbage, cauliflower, broccoli, Brussels sprouts, Chinese cabbage and related vegetable crops. Furthermore, in some countries (e.g. Japan; Dixon 1988), values of horticultural land have diminished because of clubroot infestations. The costs of control of the disease are usually very high, and often involve extensive rotations or expensive chemical applications (see below).

3.3 Symptoms

The first symptom of clubroot in brassica crops is the wilting of leaves of infected plants, especially on warm days, but the plants appear to recover overnight. As the disease progresses, leaves of infected plants turn yellow and the plants become stunted. Infected plants may survive for the life of a crop, but are unlikely to produce marketable vegetables. In severe cases, infected plants wither and are eventually killed by the disease (Karling 1968; Biggs 1994).

The wilting of the foliage and eventual death of infected plants is the direct result of damage to their root systems caused by P. brassicae. Infected roots become distorted, form galls and in severe cases become very enlarged and swollen. The swollen roots are the characteristic symptom of the disease. Heavily infected roots eventually decay. Root damage reduces the capacity of plants to obtain nutrients and water from the soil (Karling 1968; Biggs 1994).
In 1875, Woronin, working in Russia, recognised that clubroot was caused by a pathogen, which he formally named *Plasmodiophora brassicae* (Karling 1968). Before 1875, the disease was thought to have a number of possible causes, including worms, insects, soil becoming depleted by continual cultivation, over-manuring of soil, and adverse weather (Karling 1968). *Plasmodiophora brassicae* has been considered for many years to be a primitive fungus (Karling 1968). Modern taxonomic opinion places the pathogen in the Proteotista, a group of organisms, distinct from fungi, that includes all organisms that are non-plant, non-animal and non-fungal (Margulis 1990).

*Plasmodiophora brassicae* is closely related to *Spongospora subterranea*, the organism that causes powdery scab of potatoes. Both organisms have multinucleate plasmodia that develop into either sporangia, which produce zoospores, or cystosori, which form resting bodies (Dylewski 1990).

There are two phases in the life cycle of *P. brassicae*. The first involves the infection of root hairs by primary zoospores (Mithen & Magrath 1992). The zoospores attach to root hair cell walls and penetrate them using a specialised infection apparatus (Dylewski 1990). The protoplast of each zoospore is then forced into the root hair cell where it begins to enlarge at the expense of the host cells. A multilobed sporangium in which secondary zoospores develop forms later. Secondary zoospores are released at maturity into the soil, and can reinfest other healthy root hairs (Naiki et al. 1984).

The second phase of the life cycle involves development of secondary plasmodia within host root cortices and steles. The secondary plasmodia lead eventually to development of the characteristic root galls, within which cysts (resting spores) develop. The spores are released into the soil when infected plants die and decay (Karling 1968; Mithen & Magrath 1992), and these can survive for many years.

The link between the two phases of the *P. brassicae* life cycle has only been recently established. It had been suggested, though never observed, that secondary zoospores developed into amoebae and plasmodia which then migrated from root hairs into the root cortex (Karling 1968). Mithen & Magrath (1992), studying *P. brassicae* infection of *Arabidopsis thaliana* (mouse ear cress), found, however, that the early stages of infection in this plant were the same as that reported in brassicas. They observed that infection of the root cortex and subsequent formation of galls did not depend on secondary zoospore infection as previously thought. They observed small plasmodia and myxamoebae within root cortex tissue at the same time that secondary zoospores were being formed. The pathogen may produce enzymes that can digest cell walls, which would account for the observed migration of plasmodia and myxamoeba. Secondary plasmodia are thought to arise from the enlargement of small myxamoeba within host cytoplasm.
Resting spores of *P. brassicae* are easily dispersed on footwear, machinery, and contaminated and infected transplants, in water, or by livestock that have been feeding on contaminated brassicas (Karling 1968; Crute et al. 1980; Datnoff et al. 1984). Resting spores of the pathogen can also remain viable in the soil for up to 20 years (Karling 1968; Biggs 1994). Another factor that probably aids in the perpetuation and spread of the disease is the ability of *P. brassicae* to infect many alternative hosts, including cultivated and wild cruciferous plants, and a number of non-cruciferous weeds. Karling (1968) reported that more than 300 species and varieties in 61 genera of the Cruciferae were susceptible to *P. brassicae*, and that nine species of non-cruciferous plants had been found with *P. brassicae* zoosporangia in their root hairs.

There are no reports in the scientific literature that clubroot of brassicas is a seedborne disease, although Karling (1968) noted that seeds can occasionally carry *P. brassicae* resting as surface contaminants.
5 CONTROL

Because of the effects of clubroot on vegetable production, control of the disease has always been important wherever the disease has occurred. Adequate control has, however, been difficult to achieve, mainly because resting spores of *P. brassicaceae* can remain viable in soils for several years (Karling 1968). Modern intensive farming practices have also contributed to the difficulty of controlling the disease. Continual cropping of land with brassicas causes *P. brassicaceae* inoculum to build up in soil, and increases the incidence and severity of clubroot.

Over the last 20 years, considerable research effort has been expended studying clubroot of brassicas, mostly aimed at finding suitable methods to control the disease. Control methods that have been developed include use of suitable cultural practices, fertilizers, chemical treatments active against *P. brassicaceae*, and the breeding of new brassica cultivars that are resistant to clubroot.

5.1 Cultural control

Cultural methods for control of clubroot have been advocated for many years (Karling 1968). The methods suggested include use of crop rotation, leaving land fallow for several years, ensuring that there is good drainage, removing all weeds likely to act as alternative hosts of *P. brassicaceae*, and preventing animals from grazing clubroot-infected vegetable brassica crops.

These methods can help reduce the build-up of *P. brassicaceae* inoculum in soil, but will not eradicate the disease once it is established. Crop rotation or leaving land fallow for long periods is often not feasible in modern intensive vegetable cropping, and, as in Japan, there may not always be enough clubroot-free land available for adequate crop rotations (Dixon 1988). Furthermore, rotations with non-brassica crops must be long, because short rotations with crops such as ryegrass and sorghum that are not affected by *P. brassicaceae* do not reduce levels of the disease (Karling 1968; Yamagishi 1987; Harling & Kennedy 1991). The use of bait crops (see Biological control, below) has recently shown promise for clubroot control.

The removal of all cruciferous and non-cruciferous weeds from brassica crops should be part of normal cultivation practice, because alternative hosts may harbour clubroot infection. Removal of these plants should reduce the potential for spread of the disease, if it is already present. If the disease is not present then removal of these weeds will help to reduce the likelihood of clubroot becoming established.
5.2 Fertilizers

5.2.1 Lime

Applying mineral fertilizers that contain calcium (lime) to soil before planting is probably the oldest method used for clubroot control (Karling 1968; Webster & Dixon 1991a), and remains one of the commonest control methods used. The level of control achieved by liming has, however, varied from excellent to negligible. The variation in efficacy is probably related to several factors including the type and particle size of lime, soil type, *P. brassicae* inoculum level, type of additional fertilizers and chemicals applied, timing of lime applications, and prevailing climatic conditions (Karling 1968; Hamilton & Crête 1978; Dobson et al. 1983; Campbell et al. 1985).

It has long been thought that increasing the soil pH to 7.2 or greater will control clubroot. There is obviously a range of soil pHs that can give control, depending on the type of lime fertilizer used and the soil type. Furthermore, if the soil is heavily infested with *P. brassicae*, raising soil pH alone may not control the disease (Webster & Dixon 1991a).

Growth of brassica crops and the resulting yields may also be affected by liming (Karling 1968; Hamilton & Crête, 1978). Glasshouse experiments with cabbage (Hamilton & Crête 1978) have shown that calcium hydroxide added to an organic soil increased the soil pH. An increase in dry matter yield was obtained with pHs up to pH 5.2, but higher pHs reduced yields. Increasing the pH to above 7 reduced the incidence of clubroot. When calcium hydroxide was added to mineral soil the incidence of clubroot increased up to pH 6.2, then declined until no clubroot was found at pH 8.04. The pH required for maximum yield was lower than that required to control clubroot. When magnesium hydroxide, magnesium carbonate or calcium sulphate were added to the mineral soil there was a decrease in disease of the cabbages but the magnesium compounds were harmful to seed germination.

Fletcher et al. (1982) recorded a reduction in clubroot and an associated increase in marketable yield of cabbage after calcium carbonate, calcium sulphate and sodium carbonate were added to mineral soils in field experiments. Calcium carbonate raised the soil pH from 6.7 to 7.9, sodium carbonate raised the pH to 8.3 and calcium sulphate slightly reduced the pH to 6.6. Tate & Cheah (1983) demonstrated that a combination of high soil pH (7.0) and the fungicide zineb reduced clubroot and increased yield in cauliflower.

The mode of action of lime and soil pH on *P. brassicae* is not fully understood. It was originally thought that the increase in pH and the increase in the calcium ion concentration depended on each other and that both factors in combination affected *P. brassicae* populations. It has, however, been shown that both factors can act independently and together against the pathogen (Fletcher et al. 1982; Campbell et al. 1985; Webster & Dixon 1991a). Laboratory tests have indicated that liming and
increasing pH act on *P. brassicae* in two ways (Dixon & Webster 1988a; Myers & Campbell 1985); increasing either pH or calcium ion concentration, suppresses infection of host root hairs; increase in pH limits gall formation on roots. Lime probably alters the balance of available nutrients in soil to the detriment of *P. brassicae* (Campbell et al. 1985; Dixon & Webster, 1988).

The distribution of lime through soil and the particle size of the fertilizer used have been shown to affect the level of clubroot control obtained (Dobson et al. 1983). Small particle size gives more even fertilizer distribution than large particle size. Dobson et al. (1983) suggested that the contradictory results obtained when lime has been used for the control of clubroot may result from different distributions of the lime through the soil and from the usual method of obtaining soil pH from bulked soil samples. More accurate indications of the soil pH would be obtained from testing several or many individual soil samples.

Sun & Huang (1985) showed that a formulated soil amendment, containing rice husks, bagasse, oyster shell powder, urea, potassium nitrate, calcium superphosphate and mineral ash, incorporated at 0.5 and 1%, reduced clubroot in Chinese cabbage. The effect probably resulted from the high concentration of calcium in the amendment (Yang & Hsieh 1985).

5.2.2 Calcium cyanamide

The nitrogenous fertilizer calcium cyanamide, which has liming, herbicidal and fungicidal properties, has been used to control clubroot of brassicas for 60 years (Karling 1968; Dixon & Williamson 1984; Williamson & Dyce 1989). The compound has been widely used in countries such as France, Germany, U.K., U.S.A., Russia, and Japan (Dixon & Williamson 1984) and Czechoslovakia (Zvára 1981).

As with lime, the mode of action of calcium cyanamide against *P. brassicae* is not fully understood, even after extensive laboratory and field research. Laboratory experiments have shown that resting spore germination is suppressed when this compound is applied to infected Chinese cabbage seedlings (Dixon et al. 1987; Naiki & Dixon 1987). The breakdown products of calcium cyanamide are thought to affect *P. brassicae* in several different ways. Release of calcium hydroxide raises pH and calcium ion content of soil (Williamson & Dyce 1989). Urea is thought to be toxic to *P. brassicae*. The increase in plant growth and vigour from added nitrogen probably assist plants to overcome attack by *P. brassicae* (Dixon et al. 1987; Williamson & Dyce 1989).

Studies in Scotland showed that calcium cyanamide reduced clubroot symptoms in *Brassicaoleracea* by 30% (Dixon & Williamson 1984). Further studies with cabbage and cauliflower confirmed this result, and demonstrated that the compound applied to soil over several growing seasons reduced incidence and severity of the disease (Humpherson-Jones et al. 1992). Increased yields after calcium cyanamide application have been reported even for soils that are heavily infested with *P. brassicae* (Humpherson-Jones et al. 1992). A field trial in Levin, New Zealand, in 1987
demonstrated that calcium cyanamide added at 200 or 300 kg ha\(^{-1}\) to severely infested soil reduced clubroot incidence and increased yield of cauliflower by 50% (L.H. Cheah personal communication).

Although calcium cyanamide alone gives very good control of the disease, in combination with a pesticide it can give enhanced clubroot control. Mappes et al. (1989), in field trials in Germany, demonstrated that, although calcium cyanamide and the soil fumigant dazomet (Basamid\(^{®}\)) each reduced the level of disease, control was enhanced when both materials were used in combination. In one trial with Brussels sprouts where \(P.\ brassicae\) inoculum levels were low, the two materials together gave 100% control of clubroot. In another trial with higher inoculum levels, incidence of the disease was reduced from 100% with no treatment to 57% with dazomet, 68% with calcium cyanamide, and 40% with both compounds in combination. In a third trial, marketable yield of kohlrabi was increased over untreated plots by 45% with dazomet, 59% with calcium cyanamide, and 87% with both compounds together.

### 5.3 Soil fumigation and solarisation

Sterilisation of soil with chemicals such as formaldehyde, methyl bromide, dazomet, mercury compounds, chloropicrin, and pentachloronitrobenzene, has been used for many years to reduce severity and incidence of clubroot, but with varying success (Karling 1968; White & Buczacki 1977; Porter et al. 1991). Soil fumigation is an expensive method of clubroot control, however, and all of the fumigation chemicals are highly toxic to humans. Several of the chemicals are no longer used because of high toxicity, residue, or environmental problems.

Reducing pathogen inoculum in soil using solar radiation is an alternative that has been used successfully to control a number of soilborne pathogens, including \(P.\ brassicae\) (Porter & Merriman 1985). Soil solarisation has been tested for clubroot control in Japan, U.K., California and Australia, with varying degrees of success (Myers et al. 1983; Porter & Merriman 1985; Porter et al. 1991). With this method, field areas to be treated are covered with sheet plastic and left for specific periods, determined by soil temperatures achieved and the amount of solar radiation experienced (Myers et al. 1983). This method does not sterilise the soil, but inoculum levels can be reduced to levels where diseases are controlled.

Soil solarisation for clubroot control has given variable results. In the United Kingdom the method was ineffective because solar radiation levels were too low (Myers et al. 1983; Porter et al. 1991). Solarisation has also been tested in the Salinas Valley in California where clubroot has become well established (Myers et al. 1983), but was not feasible because the length of time required to achieve clubroot control (70 days) interfered with the growing cycles of brassica crops. In Victoria, Australia, Porter & Merriman (1985) found that soil solarisation reduced clubroot in broccoli and increased marketable yield. Further trials (Porter et al. 1991) have demonstrated that the method can be used to control clubroot in cauliflowers and Chinese cabbage.
They also showed that solarisation combined with low application rates of dazomet and methyl bromide soil fumigants gave good control of clubroot and increased yield. Soil type and *P. brassicae* inoculum levels influenced the effectiveness of the treatments. Less control and lower yields were obtained in heavily-infested soils than where infestation was light.

Soil moisture was found to be important in determining the length of time required to kill the *P. brassicae* resting spores using elevated soil temperatures (Porter et al. 1991). When dry soil was heated continuously in an oven at 40°C, resting spores were still viable after 28 days. No viable spores were detected after 14 days in soil at field water-holding capacity held at the same temperature.

### 5.4 Pesticides

A wide range of chemicals, including fungicides, herbicides, surfactants, trace elements and experimental compounds, has been tested for efficacy against *P. brassicae* and for control of clubroot, in both glasshouse and field experiments. Varying degrees of control have been achieved. Some compounds that have shown promise in glasshouse experiments have not been effective in field trials (e.g. Vanachter et al. (1985) with fosetyl-Al). Compounds that have given good control of clubroot include; benomyl, captafol, chlorothalonil, dichlorophen, fosetyl-Al, thiabendazole, thiophanate-methyl, thiram, tolclofos-methyl, trichlormid, boron, sodium tetraborate, and sulphur (Karling 1968; Kroll et al. 1984b; Dixon & Wilson 1984a, b; Vanachter et al. 1985; Doyle & Clancy 1986a, b; Dixon et al. 1987; Humpherson-Jones 1993). Of these, only benomyl (Benlate®), thiophanate-methyl (Topsin®), and a mixture product of chlorothalonil plus thiophanate-methyl (Taratek®) are registered in New Zealand as transplant and soil drench treatments for clubroot control (O’Connor 1994).

#### 5.4.1 Fungicides as soil drenches

The pesticides that have been most widely tested and which have good efficacy for clubroot control include benomyl, captafol, chlorothalonil, thiophanate-methyl, tolclofos-methyl, trichlormid, and zineb (Tate 1977; Tate & Eales 1982; Tate & Cheah 1983; Dixon & Wilson 1984a, b; Doyle & Clancy 1986a, b; Kroll et al. 1984b; Vanachter et al. 1985; Ohmori et al. 1986; Naiki & Dixon 1987; Rod 1992). Most efficacious fungicide treatments have been applied as soil drenches or as drench applications into seedling transplant holes.

Benomyl has been tested extensively for activity against *P. brassicae* and has been found to significantly reduce clubroot in both glasshouse and field trials in most instances (Dixon & Wilson 1984b; Kroll et al. 1984b; Vanachter et al. 1985; Naiki & Dixon 1987). Chlorothalonil has been shown to give good control of the disease (Vanachter et al. 1985). Thiophanate-methyl and tolclofos-methyl have also been found to give similar levels of clubroot control to benomyl, with concomitant yield increases (Dixon & Wilson 1984b; Doyle & Clancy 1986a, b). Thiophanate-methyl
has, however, been found to be phytotoxic to cabbage and cauliflower seedlings when applied to compost blocks before transplanting (Ann et al. 1987).

Trichlamide (NK 483; Hataclean®) is one of the most promising compounds tested recently for control of clubroot. The chemical was developed in Japan in the late 1970s as a broad-spectrum fungicide active against a variety of soil-borne pathogens (Dixon 1988), and has been successfully tested against *P. brassicae* (Ohmori et al. 1986; Dixon 1988). Trichlamide was developed as an alternative to quintozene (pentachloronitrobenzene; PCNB), which was extensively used in Japan for clubroot control, but which has been phased out for environmental and human health reasons. Suitable chemical control of clubroot is necessary in Japan because the limited horticultural land suitable for brassica production must be very intensively cropped, making cultural, and fertilizer methods of clubroot control unsatisfactory.

Trichlamide has been tested in the UK and Belgium in glasshouse and field trials (Dixon & Wilson 1984a, b; Vanachter et al. 1985; Doyle & Clancy 1986b). In glasshouse trials, the compound reduced both the incidence and severity of clubroot in Chinese cabbage and cauliflower. When tested under field conditions in Scotland, trichlamide significantly reduced clubroot expression on cabbage and the yield increased (Dixon & Wilson 1984a, b). Vanachter et al. (1985) did not however, get comparable results with trichlamide in field trials with Chinese cabbage and cauliflower in Belgium. They achieved only a small reduction in disease in soils heavily infested with *P. brassicae*.

Humpherson-Jones (1993) has recently demonstrated that dichlorophen-Na (Panacide®; Mostox®) and fluazinam (PP-192; Shirlan®), a compound originating from Japan (Worthing & Hance 1991), controlled clubroot in cabbage. Recent results in Australia have also demonstrated efficacy of this compound for clubroot control (ICI confidential reports; personal communication D.M. Collins & P.A. Taylor). Flusulphamid (Nebijin®), another compound developed recently in Japan, is also effective against *P. brassicae* (Worthing & Hance 1991). Control of powdery scab of potatoes, which is caused by a pathogen closely related to *P. brassicae*, has recently been achieved with dichlofluanid (Euparen®), dichlorophen-Na, flusulfamide, fluazinam, and mancozeb (Braithwaite et al. 1994; Falloon et al. 1994; S.J. Wale personal communication). These recent results suggest new and novel fungicides should continue to be tested against clubroot of brassicas. Several candidate compounds, particularly those originating from Japan (e.g. flusulfamide, fluazinam, trichlamide), may be worthwhile alternative chemicals for clubroot control.

### 5.4.2 Fungicide transplant dips
Dipping of brassica transplants in solutions or slurries of efficacious fungicides has also been tested for clubroot control. This method of application has usually been less effective than soil drench application (e.g. Tate 1977 with benzimidazoles).
5.4.3 Fungicide seed treatments
Benomyl, thiram, thiophanate-methyl and several other fungicides were tested as seed treatments for control of clubroot by Rod (1992). Seed of Chinese cabbage was mixed with different rates of fungicides before sowing, then grown under optimal conditions for five weeks. Statistically significant reductions in clubroot were observed with benomyl, thiram and thiophanate-methyl seed treatments, but the reductions were small, and the fungicide rate required (20 g/kg seed) was toxic to seedlings, suggesting that seed treatments alone are unlikely to be useful for control of clubroot. The small amount of chemical applied on seed may not be enough to control infection of plants from soilborne inoculum beyond the seedling stage.

5.4.4 Trace elements
Doyle & Clancy (1986a) found that when thiophanate-methyl was used in combination with boron or sulphur, level of clubroot control and increases in yields were greater than those obtained with thiophanate-methyl alone. Of the combinations, sulphur plus thiophanate-methyl resulted in the lowest number of diseased plants, the highest yield and a very low score for disease severity of all the products tested. Tolclofos-methyl was less effective in controlling clubroot in comparison to thiophanate-methyl. Boron or sulphur alone did not reduce disease incidence or severity. Exactly how boron and sulphur increase the effectiveness of thiophanate-methyl is not known, although Webster & Dixon (1991b) found that boron may affect the maturation rate of *P. brassicae* root hair infections and the development of galls.

5.4.5 Other pesticides
Herbicides and insecticides are commonly used for weed and pest control in brassica crops, but little is known about their effects on clubroot. Doyle & Clancy (1986a) tested the herbicides trifluralin and napropamid alone and in combination with trace elements and fungicides for effects on clubroot control. The herbicides alone did not affect the disease, but when trifluralin was applied in combination with thiophanate-methyl, there was a significant reduction in disease. When the herbicide was applied in combination with boron or sulphur, there was also a statistically significant reduction in disease incidence but it did not affect yield. The reductions were not as great as those recorded for thiophanate-methyl. McIntosh et al. (1983), reporting a glasshouse experiment, demonstrated that some phenoxy acetic acid herbicides have activity against *P. brassicae*. Humpherson-Jones (1993) has recently demonstrated that the surfactants sodium dioctyl sulphosuccinate and alkyl phenyl ethylene oxide have activity against clubroot of cabbage. These results suggest that it would be worthwhile to test non-fungicide pesticides that are, or could be, used on brassica crops for efficacy against clubroot.

Chlorine (Cl) has been tested for control of *P. brassicae* contamination in irrigation water used in southwest Virginia on brassica transplant seedbeds which had not previously experienced clubroot infection (Datnoff et al. 1984). The source of contamination was from runoff from fields infested with the pathogen, and cabbage
seedlings grown in sediment from irrigation ponds developed clubroot. In laboratory tests, 2 mg l⁻¹ in water, after 5 min exposure, was effective in reducing clubroot in cabbage, while field trials demonstrated that 200 mg l⁻¹ Cl added to irrigation water reduced clubroot incidence, but also reduced plant growth and yield (Datnoff et al. 1987).

5.5 Disease resistance

The search for resistance to clubroot, and the breeding of resistant brassica cultivars, is becoming more important as the traditional methods of clubroot control are becoming less acceptable or effective. While the concept of selecting appropriate plants that show resistance to *P. brassicae* and introducing them into breeding programmes is simple, achievement of useful and stable resistance has been difficult. *Plasmodiophora brassicae* exists as a number of pathotypes/races which makes it difficult to identify which form(s) particular breeding lines are resistant to (Williams 1966; Crute et al. 1980). The genetic control of resistance to *P. brassicae* in brassicas is incompletely understood, and there is little information on the genetics of *P. brassicae* (Crute et al. 1980; Crute 1986).

The differential pathogenicity of *P. brassicae* has been known for over 70 years (Crute et al. 1980), but it was not until the late 1950s that a series of differential hosts was developed that enabled identification of different *P. brassicae* pathotypes (Williams 1966; Crute et al. 1980). Researchers in different areas of the world began using differential hosts to identify the pathotypes of *P. brassicae* present, but different sets of differential hosts were used, making comparison of data very difficult. This also made it difficult for plant breeders to determine which pathotypes to use for screening of their breeding material (Williams 1966; Buczacki et al. 1975).

Buczacki et al. (1975) developed a standard group of differential hosts in an effort to overcome the problems associated with multiple identification systems for *P. brassicae* pathotypes. The group, designated the European Clubroot Differential (ECD) Set, includes 15 hosts, five each from *Brassica campestris*, *B. napus*, and *B. oleracea*. Some of the hosts chosen had been commonly used by previous workers in their differential host sets (Buczaki et al. 1975; Crute et al. 1980). Each differential is designated by a number which is used to assign an overall code to the specific *P. brassicae* race being tested, depending on the reactions of the hosts.

There are several drawbacks with the ECD host set. One is that not all of the differentials are commonly used in brassica breeding programmes (Crute et al. 1980), making it difficult to identify resistance factors, if any, that are present in breeding stock when similar resistance factors and genes may not be present in the host series. Furthermore, not all the theoretical and possible outcomes of host and pathogen interaction can be accommodated by some of the hosts (Crute et al. 1980). The exact number of genes involved in the observed resistance to *P. brassicae* in each of the three host groupings is also not completely known. An example of this is seen in
the *B. napus* hosts, where resistance was thought to be governed by three genes. However, two pathotypes of *P. brassicae* have been identified, but these would not have occurred if only three genes were involved. Crute et al. (1980) suggested that a minimum of five genes is involved in the resistance of the *B. napus* ECD hosts.

The number of genes involved in resistance in *B. oleracea* is also uncertain (Crute et al. 1980; Crute 1986). Resistance in the cabbage cv. ‘Badger Shipper’ is thought to be recessive and under polygenic control, but the exact number of genes involved is not known. Resistance in the broccoli line ‘MSU 134’, developed in North America, is thought to be either under the control of two genes, one recessive and one incompletely dominant, or controlled by a single recessive gene (Crute et al. 1980).

Crute et al. (1983) studied the relationship between *B. oleracea*, *B. napus*, *B. campestris* and *P. brassicae* and found that resistance in *B. napus* and *B. campestris* was differential, i.e. both cultivar and pathogen exhibit variation and governed by several major genes. Resistance in *B. oleracea* was found to be non-differential i.e. one cultivar is always more resistant irrespective of the pathogen and one pathogen is always more virulent regardless of cultivar genotype. It is also possible that the variation in resistance in *B. oleracea* results from environmental factors and the population density of *P. brassicae* (Crisp et al. 1989).

Little is known about the genetics of *P. brassicae*; several different pathotypes of the pathogen have been recorded, it is difficult to identify particular pathotypes, and it is not known if individual pathotypes are stable (Crute 1986). The life cycle of *P. brassicae* is still not adequately understood, and the pathogen is obligate (cannot be cultured on artificial media). These factors make cloning and maintenance of stable pathogen lines on live hosts difficult.

Even though fundamental knowledge on the genetic control of resistance to *P. brassicae* in brassicas is lacking, resistance has been found and new resistant cultivars are being developed and released commercially (Crute et al. 1980; Dixon 1988; Yamagishi 1987; Chiang & Crête 1989; Hansen 1989). Many extensive screening programmes have been carried out in efforts to identify resistant material from worldwide acquisitions of germplasm (Yoshikawa 1983; Yamigashi 1987; Crisp et al. 1989; Bradshaw & Williamson 1991).

From these screening programmes, it is now known that resistance to *P. brassicae* is common in *B. campestris*, especially in turnip varieties, but not in Chinese cabbage, and in *B. napus*, especially in the swede and rutabaga varieties. Resistance is not common in *B. oleracea* (Crute et al. 1980; Yoshikawa 1983; Yamagishi 1987; Crisp et al. 1989). Lack of resistance in *B. oleracea* has led several groups of workers to carry out extensive screening programmes concentrating on this species (Dixon & Robinson 1986; Crisp et al. 1989; Bradshaw & Williamson 1991).

Crisp et al. (1989) screened about 1000 *B. oleracea* accessions for resistance to *P. brassicae*, and showed that kales are least susceptible to the pathogen, kohlrabi,
cauliflower and broccoli are the most susceptible, and cabbage and Brussels sprouts vary in susceptibility. The kales showed significant variation both within and between types irrespective of their origins. All landrace types of broccoli, cauliflower, and cabbage from Italy showed lower levels of susceptibility and greater variation within their own type than was found from those types originating outside Italy. A new source of resistance was identified in Irish landrace cabbages. These cabbages also showed significant variation within type and could possibly be used in future clubroot resistance breeding programmes.

Of all the accessions screened by Crisp et al. (1989) in Britain, the 30 accessions showing the lowest levels of susceptibility to *P. brassicae* were cabbages, kales, or derivatives of these. The cabbage types included Böhmerwaldkohl, originating from the Germany-Czechoslovakia border, and Bindsachsener, from central Germany. Both types have been used extensively in breeding programmes throughout the world. Böhmerwaldkohl was used to develop the clubroot resistant cabbage cv. 'Resista' (Crisp et al. 1989; Hansen 1989). Resista has since been withdrawn in Norway because of its poor performance (Hansen 1989). Bindsachsener is thought to have been used in the breeding of the North American clubroot-resistant cabbage cvs 'Oregon 100', '123', '140' and '142' (Crisp et al. 1989).

Intra- and inter-specific hybridization is being used as a means of breeding clubroot-resistant cabbage, broccoli, cauliflower, and other brassica varieties. The resistant cabbage cv. 'Badger Shipper' is an example. The resistance in this cultivar originated from a chance kale x cabbage hybrid, and is known as the "Wisconsin" source (Crute et al. 1980). This source has also been used to develop resistance in Brussels sprouts, calabrese-broccoli, and cauliflower varieties (Crute et al. 1980; Crute 1986; Crisp et al. 1989). Two resistant broccoli lines, 'CR-1' developed by Oregon State University and 'MSU 134', are also thought to have originated from the Wisconsin source (Crute et al. 1980).

Japanese brassica breeders, after demonstrating that local varieties were all susceptible to *P. brassicae*, have tried to exploit European sources of resistance in extensive breeding programmes (Yoshikawa 1983; Yamagishi 1987; Dixon 1988). The first programme began in 1974 to develop a clubroot-resistant Chinese cabbage (Yoshikawa 1983). Susceptible Chinese cabbage was crossed with a resistant European fodder turnip. Resistance was controlled by a single gene which was independent of leaf character which made it simple to recover the desired resistance character. This programme has resulted in several clubroot-resistant Chinese cabbage, turnip and salt green lines being released commercially (Yamagishi 1987; Dixon 1988).

A breeding programme for clubroot resistance in cabbage has also been instigated in Japan, but stable resistance in progeny have been difficult to obtain. The resistance source used was the European cabbage Böhmerwaldkohl. One variety of resistant cabbage has, however, been developed and released commercially in Japan (Yoshikawa 1983; Yamagishi 1987). More success has been achieved using kale as
a source of resistance. Kale has a greater level of resistance than Böhmerwaldkohl and resistance is inherited by a single recessive gene (Yoshikawa 1983). Several clubroot-resistant calabrese varieties have been released commercially (Dixon 1988), and cabbage, broccoli, cauliflower, and Brussels sprouts types are being developed (Yoshikawa 1983).

A clubroot-resistant cabbage cv. 'Richelain' that has multi-race resistance to three *P. brassicae* pathotypes has been developed in Canada (Chiang & Crête 1989). This variety was developed from an interspecific cross between a clubroot-resistant rutabaga and a colchicine-induced tetraploid cabbage. Clubroot-resistant cabbages that are low in glucosinolates have also been developed in Canada (Chong et al. 1981; Chiang et al. 1984). Glucosinolates and their breakdown products give brassicas their characteristic flavour and taste but are known to have toxic properties and to possibly inhibit growth. It is thought that the susceptibility of some crucifers to clubroot is related to the presence of certain glucosinolates. Some of the glucosinolates release high levels of auxins which have been associated with the abnormal growth of roots infected with *P. brassicae* (Chong et al. 1981; Chong et al., 1985). When a number of commercially available cabbage lines and clubroot-resistant breeding lines were analysed for the presence of the thiocyanate ion, it was found that resistant cabbages contained significantly less of this product than did the commercially available cultivars. Several of the resistant breeding lines were found to be low in isothiocyanate and goitrin and some lines were free of goitrin. There is a possible association between the consumption of cruciferous crops and the development of goitre, so the breeding of clubroot-resistant varieties that are low in goitrin would be very desirable (Chiang et al. 1984; Chong et al. 1985).

Although there have been a number of successes in breeding clubroot-resistant brassicas, several difficulties have also been encountered. If interspecific crosses are used to obtain resistance, it may be difficult to breed the desired vegetable quality characters into cultivars, because some of the genes involved in resistance are linked to these characters (Yoshikawa 1983). Male sterility can also cause problems with interspecific crosses (Chiang et al. 1984). Heterogeneity in *P. brassicae* may also cause problems. In many countries, multiple races of *P. brassicae* have been recorded; seven have been recorded in Japan (Yoshikawa 1983), six or seven in Canada (Crute et al. 1980), nine in the USA (Williams 1966), and eight have been differentiated in New Zealand, mainly on forage brassica cultivars (Lammerink 1986).

Resistance may also break down over time with continuous cropping. This has occurred with the cabbage cv. 'Badger Shipper' (Crute *et al*. 1980; Crute 1986), probably because of the build-up of a more pathogenic population of *P. brassicae*. Resistance in this cultivar was restored after a break of three years because of the removal of selection pressure for the more virulent population.

Gabrielson & Robak (1988) have demonstrated that temperature can affect the level of resistance observed in cultivars. In glasshouse tests, it was shown that the higher the temperature the more susceptible the plants were to clubroot. Badger Shipper
cabbage, a resistant cauliflower, and broccoli were resistant to one pathotype of *P. brassicae* (pathotype 6) at 15°C and 20°C and only partially resistant at 25°C and 30°C, while they were resistant to a second pathotype at 15°C and very susceptible to it at 20°C and over. Temperature sensitivity of resistance could explain why some breeding lines show clubroot resistance in the field but not under optimum disease conditions in glasshouse tests.

### 5.6 Biological control

The possibility of controlling clubroot by using bacteria and/or fungi that are antagonistic to *P. brassicae* has not been extensively investigated. Djanika (1991) in Indonesia has isolated a fungus from soil infested with *P. brassicae* that reduced clubroot in glasshouse and field trials, but only in non-limed soil. Kroll et al. (1984a) reported that five of six root surface-inhabiting bacterial strains reduced incidence of clubroot on radish plants in a growth room experiment. The bacteria had little effect on root hair infection but reduced colonization of root steles by *P. brassicae*. No effect was detected in a field trial which tested one of the isolates.

The use of bait crops for control of clubroot has been investigated. This approach uses a crop susceptible to *P. brassicae* which is grown for a short time (a few weeks), then destroyed before planting the desired vegetable brassica crop. The soil inoculum levels are reduced by stimulating resting spore germination, which results in reduced clubroot in the subsequent vegetable brassica crop. Yamagishi (1987) reported success in controlling clubroot when turnip and kale resistant to *P. brassicae* were grown before a main crop of either Chinese cabbage or turnip, both of which were susceptible to the disease. Harling & Kennedy (1991) found that growing a bait crop of oilseed rape before growing calabrese helped to control clubroot, and this was particularly efficacious when used in conjunction with liming.
CONCLUSIONS

Clubroot of vegetable brassicas has long been recognised as a harmful disease which occurs wherever susceptible crops are grown. Many different approaches to control of the disease have been investigated. Control of clubroot has been obtained using various cultural methods, fertilizers, pesticide chemicals, and resistant cultivars. The ability of *P. brassicae* to infect many different plants, to produce large numbers of highly resistant resting spores, and to survive for many years, make the disease difficult to control (Karling 1968). The use of resistant cultivars, only a few of which are commercially available, probably offers the best prospect for effective and economic clubroot control. Integrated control of the disease, using a combination of cultural, chemical, disease resistance, and possibly, in the future, biological strategies, has been widely advocated (e.g. Yamagishi 1987; Harling & Kennedy 1991). Vegetable brassica growers in New Zealand should re-acquaint themselves with the available methods to alleviate clubroot, and take a fully integrated approach to control of the disease. Research on the disease should concentrate on pesticide chemicals that show promise for clubroot control, on assessment of relative clubroot resistance in currently available cultivars so that recommendations on cultivars appropriate for heavily infested areas can be made, and possibly to also investigate the potential for biological control of the disease.


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