## Occurrence of clubroot and *Plasmodiophora brassicae* Wor. races in Finland

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Abstract. Examination of clubroot in cruciferous vegetables in Finland in 1974—1978 revealed the disease in 81 % of the 101 communes inspected. The disease was most common in southern and central Finland, but was also discovered in the northern parts of the region in which cruciferous crops were cultivated (66—67° N. lat.). Clubroot was found in 65 % of the 375 plant samples collected. It occurred in 68 % of the samples of the most commonly cultivated vegetable, cabbage (56 % of the material), in 63 % of the cauliflower samples (22 % of the material), in 56 % of the samples of other cole species (13 % of the material) and in 64 % of the samples of cruciferous root crops (10 % of the material). *P. brassicae* race determinations were performed on 90 samples. The classification system of WILLIAMS (1966) was applied. The races that were isolated were 1, 2, 3, 4, 6 and 7. Race 2 was by far the most common, being found in 32 communes; races 3, 4, 6 and 7 were each found in 9—12 communes; race 1 only in one commune. No clear differences in the occurrence of the races in the various parts of the country could be observed. A comparison is made between Williams' and the ECD (BUCZACKI et al. 1975) classification systems. In addition, the pathotypes in clubroot material from Norway and Iceland were determined.

Index words: clubroot, Plasmodiophora brassicae Wor., pathotypes, races, cruciferous vegetables

#### Introduction

The records on the occurrence of clubroot disease in Finland date back to the 1860s (JAMALAINEN 1936). It is assumed that the disease came to Finland from the east, from Russia (RAINIO 1930). In his studies on the cause of crucifer hernia, WORONIN (1878) mentions that cabbage plant materials used by him came from the regions of St. Petersburg (Leningrad) and Wiborg<sup>1</sup> in Finland. According to the information collected by the Department of Plant Pathology, Agricultural Research Centre, clubroot had spread all over the country, including the western regions, by the 1930s

<sup>&</sup>lt;sup>1</sup> Since 1944 that area has been part of the Soviet Union.

(RAINIO 1930, JAMALAINEN 1936). In the 1930s the susceptibility of cruciferous vegetable cultivars and wild *Brassicaceae* plants to the fungus *Plasmodiophora brassicae* Wor. was also studied at the Research Centre. In addition, experiments for chemical control of the disease were carried out (JAMALAINEN 1936). The efficiency of new pesticides was studied in 1944—1977 (LINNASALMI 1948, 1952, 1959; LINNASALMI and TIITTANEN 1960; MURTOMAA and UOTI 1972; annual reports of the Department of Plant Pathology of the Agricultural Research Centre 1960—1977, mimeographs).

A preliminary study on *P. brassicae* races in Finland had been carried out in the years 1971—1972 (LINNASALMI and PALONEN 1974).

In this study, the occurrence of clubroot and the pathotypes of *P. brassicae* in cruciferous vegetables in Finland in 1974—1978 were examined. In addition, the pathotypes in clubroot material sampled in Norway and Iceland were determined.

The study is a part of the Nordic clubroot project, NKJ project 27 1974—1977, of the Nordic Joint Committee for Agricultural Research: Breeding for clubroot resistance, *Plasmodiophora brassicae* Wor. races, and the efficiency of new pesticides.

#### Materials and methods

#### Collection of the basic material

Most of the material for studying the distribution of clubroot was collected by inspecting cruciferous vegetable fields throughout Finland during the growing season, from June to October. A small part of the material, about 2 %, was sent in by agricultural research stations, advisers of agricultural information organizations and farmers. The inspections chiefly focused on large cole and root crop farms, but a considerable number of small farmers' crucifer fields were also inspected. The research material collected as described above can be assumed to give a general picture of the distribution of clubroot disease and the *P. brassicae* races in the period under study in the 1970s in Finland.

Samples of galls from the various cultivars affected with clubroot were taken for the analysis of *P. brassicae* races.

# Propagation of P. brassicae club material and preparation of the inoculum

The young galls of the original samples were washed thoroughly with running water. They were crushed to prepare a water suspension, which was used to inoculate a steamed (at 100°C for 1 h) soil-peat medium. From the samples for race analysis additional club material was grown on the original plant species and, if possible, on the original cultivar or on a cultivar known to be susceptible to P. brassicae, such as cabbage cv. Ditmarsk and cauliflower cv. Erfurter. To check and ensure the viability of the inocula, the black mustard (Brassica nigra (L.) W.D.J. KOCH) breeding line Sv. 72-6842 of the Swedish Seed Association, Sweden, which was very susceptible to clubroot, was also used as a host plant both during the propagation of club materials and later in identification tests of P. brassicae races.

During the initial stage of the study, the inoculation of the host was repeated two or three times as five-week growth periods. As the sampling technique improved, the club material from the first propagation could be used as basic material to grow callus cultures and small-club material. The material was stored at  $-18^{\circ}$ C.

The *callus cultures* were prepared from young galls, 1–2 cm long, which were surface-disinfected with ethanol ( $C_2H_5OH$ , 94 %) and mercuric chloride (HgCl<sub>2</sub>, 1 %) and rinsed with distilled water. Small pieces, ca. 2 mm in diameter, were cut from the inside of the galls, treated with ethanol (99.5 %) for a few seconds and rinsed with distilled water. The pieces were placed in Erlenmeyer flasks or test tubes on an agar medium. The nutrient solution used was a modification of the solution of MURASHIGE and SKOOG (1962), in which 3-indoleacetic acid had been replaced with 1-naphthylacetic acid. Coconut milk (100 ml/l) was added because our tests had shown that it increased the growth rate of callus tissue considerably. The cultures were incubated in the dark at ca. 22°C. Small pieces of callus tissue were transferred every 7 to 10 days to a fresh culture medium, the passages totalling three to four. The callus material was stored at  $-5^{\circ}$ C.

For preparing the inoculum, the callus and small-club materials were crushed mechanically, suspended in distilled water and filtered through a nylon filter cloth. The filtrate was centrifuged (2400 g, 7 min.) three to four times. Before each test, the suspension used was diluted to a concentration of 10<sup>8</sup> spores per ml, using a hemocytometer.

## Isolation and identification of races

## Method

The isolation and identification of races was carried out according to the system of WIL-LIAMS (1966), a method based on selecting four *Brassicaceae* plants as differential hosts and studying their resistance or susceptibility to *P. brassicae*.

The test plants were cabbage (*Brassica oleracea* L. convar. *capitata* (L.) Alef.), cultivars Jersey Queen and Badger Shipper, and swede (*Brassica napus* L. var. *napobrassica* (L.) Rchb.), cultivars Laurentian and Wilhelmsburger. On the basis of 16 possible interactions, 16 races can be isolated and classified according to the following scheme (WILLIAMS 1966).

The use of genetically uniform, homozygous differential host material is a prerequisite for the reliability of the race determinations. The seed material used in this study was

Possible host reactions to infection by races of Plasmodiophora brassicae.

Differential			Race														
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Cabbage:	Jersey Queen	+	+	+	+	_	+	+	_	_	+	_	+	_	_	_	_
	Badger Shipper	_	+	_	+	_	_	+	_	_	+	+	_	+	+	+	_
Rutabaga:	Laurentian	+	+	+	+	_	_	_	+	+	_	+	_	+	_	_	_
	Wilhelmsburger	+	_	_	+	_	_	_	_	+	+	+	+	_	+	_	+

+ Indicates a susceptible host reaction; - indicates a resistant host reaction.

supplied by NKJ project member Dr. R. JÖNSSON, and produced or acquired by the Swedish Seed Association (nowadays Svalöf AB), Svalöv, Sweden.

#### Testing

The tests were carried out in a glasshouse, mean temperature 22°C (20–24°C). When necessary (in winter), Osram HQL 400 W/R mercury high-pressure lamps were used for supplementary lighting.

The growing medium, Enhetsjord K (AB W. Plantin & Co, Oxie, Sweden), was a fully fertilized, fine-grained mixture of clay and

peat, pH 6.3  $\pm$  0.2. It was steamed at 100°C for 1 h. Seeds of differential hosts were sown in 9  $\times$  9 cm pots of thermosetting plastic, 10 seeds/pot, four replicates. Each pot had a plastic tray of its own. Inoculation with the spore suspension, 10 ml/pot, was performed after the sowing and the seeds were covered with a 0.5 cm layer of the growing medium. Throughout the growing period, the plants were watered by pouring water into the trays to keep the moisture even and to prevent the possibility of cross-contamination.

The growing period was from four to six weeks. The galls were well developed, the

average size being 0.5 to 1.0 cm<sup>3</sup>. They were light in colour and usually contained a great number of spores.

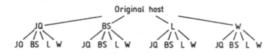
To conclude the test, the roots were washed with running water and rinsed with distilled water. The degree of clubroot infection of each plant was assessed on a scale of 0-3, and the clubroot index was calculated according to WILLIAMS (1966):

clubroot index 
$$\frac{0 \times n_0 + 10 \times n_1 + 60 \times n_2 + 100 \times n_3}{n_0 + n_1 + n_2 + n_3}$$

 $n_0 = no clubs$ 

 $n_1 = a$  few small clubs on the secondary roots  $n_2 = considerable$  clubbing on the lateral roots  $n_3 = severe$  clubs on the primary and secondary roots

This was the primary analysis. The clubs from the replicates of each differential host were combined to make a spore suspension, which was used to re-inoculate all four differential host plants, in accordance with the following scheme:



If the result of this cross-testing was similar to that of the primary analysis with respect to both degree of infection and clubroot index, the race determination could be considered to be reliable. If the result was not similar, the cross-testing was repeated three to four times.<sup>2</sup>

# Comparison with the ECD classification system

Comparison with the ECD classification system, developed by the International Clubroot Working Group (BUCZACKI et al. 1975) was performed with some of our *P. brassicae* isolates (totalling 12) classified by Williams' system. The seeds of ECD differential hosts in *Brassica campestris, B. napus* and *B. oleracea* groups (totalling 15 hosts) were received in 1976 from Dr. H. TOXOPEUS, Instituut de Haaff, Stichting voor Plantenveredeling, Wageningen, the Netherlands.

The test arrangement and the growth conditions were the same as in the tests performed with Williams' method (p. 417).

## Results and their evaluation

#### Occurrence of clubroot

## Regional distribution

Of the 192 farms, 61 %, and of the collected plant samples, altogether 375, 65 %, showed occurrence of clubroot (Table 1, Fig. 1). Thus the study revealed that clubroot was quite common all over Finland. Although the locations inspected were distributed over the country, the number of samples from the various areas differed so much that it is impossible to draw far-reaching conclusions about regional differences in the occurrence of the disease. It can, however, be noted that clubroot was fairly common in the old, densely populated farming areas, Uusimaa (N), Varsinais-Suomi (Ab) and Etelä-Häme (Ta), and also in Etelä-Savo (Sa) and Etelä-Karjala (Ka). The disease was more severe in eastern Finland than in western Finland. This was possibly partly caused by cultivation techniques, mainly by the fewer opportunities for crop rotation on the farms in eastern Finland. In these areas many farmers had to give up the production of cruciferous vegetables altogether because of clubroot disease in the 1960s and '70s.

It also turned out that clubroot was often more common, and even more severe, on smaller than on larger farms, one probable reason being the limited opportunities for crop rotation. This was evident, for example, in gardens around population centres, where var-

<sup>&</sup>lt;sup>2</sup> Purified and identified race material was sent to the Scandinavian members of this project from 1974 onwards to be used in their resistance breeding work. Since 1980 the type isolate material has been deposited in the race bank at the Swedish University of Agricultural Sciences, Department of Resistance Biology, Alnarp, Sweden.

Table 1. Distribution of clubroot and occurrence of *Plasmodiophora brassicae* Wor. races in Finland, 1974–1978.

Biological province <sup>1</sup>			mmunes		arms spected	Sa	amples	1	P. b 2	rass 3	icae 4	rac 6	es 7	1—7
		total no.	with clubroot %	total no.	with clubroot %	total no.	with clubroot %		no	. of	isol	ates		total
Ab	Varsinais-Suomi	13	100	19	100	46	100	0	6	1	2	2	1	12
Ν	Uusimaa	6	100	11	100	28	100	0	3	1	2	0	1	7
Ka	Etelä-Karjala	4	100	9	56	14	71	0	3	1	0	0	0	4
St	Satakunta	12	75	21	48	26	42	0	4	0	4	0	0	8
Та	Etelä-Häme	20	95	37	73	88	77	0	11	0	1	8	3	23
Sa	Etelä-Savo	8	50	13	54	20	55	0	1	4	0	0	1	6
Oa	Etelä-Pohjanmaa	12	58	27	37	44	32	0	2	0	2	0	2	6
Tb	Pohjois-Häme	9	56	25	24	46	24	0	1	0	1	0	2	4
Sb	Pohjois-Savo	7	100	10	80	19	84	1	1	4	0	2	0	8
Kb	Pohjois-Karjala	6	83	10	60	23	52	0	7	1	0	0	0	8
Om	Keski-Pohjanmaa	2	50	5	60	6	50	0	1	0	0	0	0	1
Ob	Pohjois-Pohjanmaa	2	100	5	100	15	100	0	0	0	0	2	1	3
tota	l/mean	101	81	192	61	375	65	1	40	12	12	14	11	90

<sup>1</sup> Heikinheimo and Raatikainen (1971).

ious cole species and cruciferous root crops had been grown in the same fields for many years.

## Clubroot in different plant species

Cruciferous vegetables are cultivated almost throughout Finland up to 66—67° N. lat., but the main production takes place in the southern part of the country. The areas of the most commonly cultivated crops in 1974—1978 (mean) are presented below (Statistics of the National Board of Agriculture (Finland).)

cabbage	cauliflower	swede
554	328	1164
118	23	733
29	10	72
701	361	1969
	554 118 29	554 328   118 23   29 10

In the 1980s, the areas of cabbage and cauliflower remained roughly the same, totalling some 1000 ha. The area of swede decreased markedly, from almost 2000 to 400 ha. The cultivation of Chinese cabbage increased to 600 ha. The total area of the other cole species was some 80 ha and that of radish and turnip totalled some 40 ha.

The data on the occurrence of clubroot in different plant species are presented in Table 2. Clubroot was found in 68 % of the cabbage samples, in 63 % of the cauliflower samples, in 56 % of the samples of other cole species, and in 64 % of the root crop samples. The distribution and number of samples from different plant species roughly indicate the frequency of cultivation of these crops.

# Plasmodiophora brassicae races and their occurrence

As in practice it was not possible, within the limits of the study, to carry out a race determination of every clubroot sample, the samples for tests were chosen to represent as many communes as possible in each biological prov-

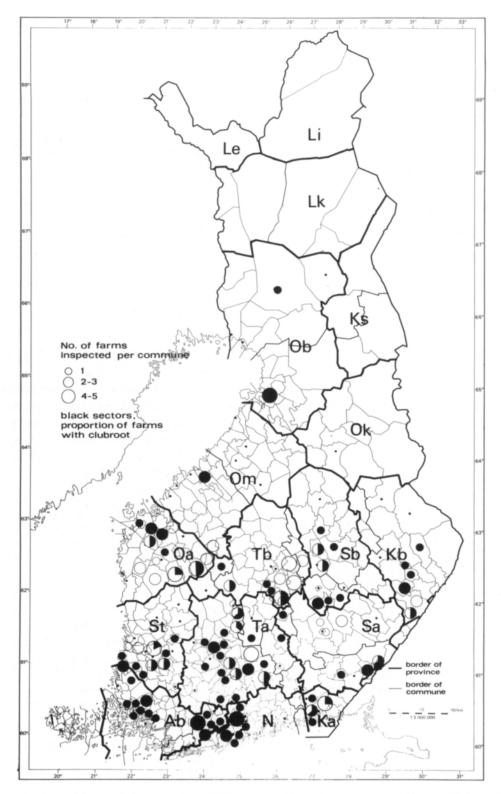


Fig. 1. Location of inspected farms growing cruciferous vegetables and occurrence of clubroot in Finland, 1974 - 1978.

Table 2. Occurrence of clubroot in different plant species.

Species	San	nples	Plant	species
	total number	with clubroot	% of samples	% with clubroot
Cabbage	209	143	56	68
Cauliflower	82	52	22	63
Broccoli	21	10		
Red cabbage	10	8		
Brussels sprouts	8	4		
Kohlrabi	3	3	13	56
Curly kale	3	1		
Marrowstem kale	2	1		
Chinese cabbage	1	0 )		
Swede	28	16		
Turnip	5	4	10	~
Fodder turnip	1	1	10	64
Radish	2	2 ]		
	375	245		

ince, altogether 69 communes, or 84 % of the communes where clubroot was found. Most determinations were performed on club samples of the crucifers cultivated most frequently: cabbage and cauliflower. Some additional determinations were made on samples taken from less common species. The number of *P. brassicae* isolates totalled 90.

## Races

The races isolated were 1, 2, 3, 4, 6 and 7 (Appendix 1). The identification results for races 1, 2, 4 and 7 were clear. The test results for races 3 and 6 showed some deviation from the classification scheme. Several isolates from both of these races showed slight contamination in Badger Shipper, although according to the scheme this differential host should have been resistant to these races.

As the identification tests with certain isolates of other races also showed more variation in the clubroot indices in Badger Shipper than in other differential hosts, it seems that the seed material of Badger Shipper may have had some genetic heterogeneity. It is also possible that the isolate materials classified as races 3 and 6 included some pathotypes that were variants of the main races. The possibility of mutation in the differential hosts or in the purified *P. brassicae* race isolates must also be taken into account.

A preliminary report on the occurrence of *P. brassicae* races determined in the present study was presented at the Brassica conference 1981 (LINNASALMI and TOIVIAINEN 1981).

#### Regional distribution

The regional distribution of the races is shown in Table 1 and on the map in Fig. 2. The communes from which the samples were taken and data on the original host are given in App. 1. Of the six races isolated, race 2 was by far the most common. It was found in 32 communes, i.e. in 46 % of the communes with clubroot. Races 3, 4, 6 and 7 were each found in about ten communes (9-12 communes, i.e. 14-17 %); race 1 occurred only once. Two different races were found in only a few communes: races 2 and 3 in Nurmijärvi (N) and Joutseno (Sa), races 2 and 6 in Haukivuori (Sb), races 6 and 7 in Hartola (Ta), Kangasala (Ta) and Oulu (Ob). On the basis of this study, no clear prevalence of races can be demonstrated in different parts of Finland. All races were distributed fairly evenly over the country, with the exception of the

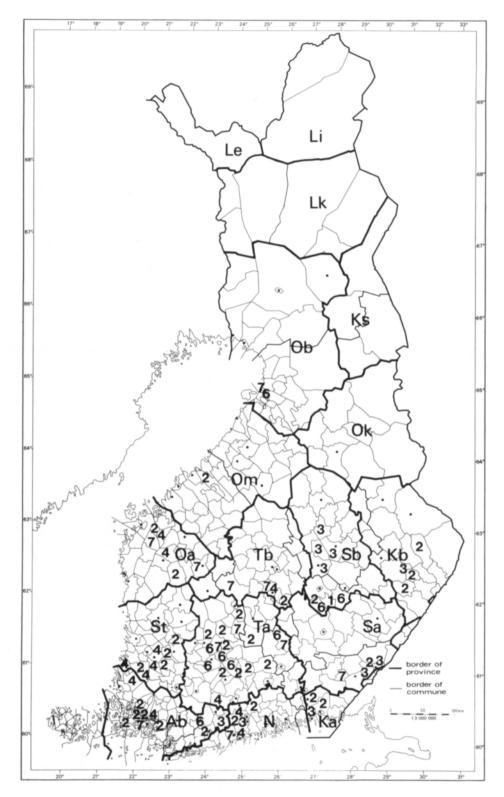


Fig. 2. Occurrence of Plasmodiophora brassicae Wor. races in Finland, 1974-1978.

rare race 1. However, race 4 seems to show some prevalence in the western parts of the country, while race 3 occurs mostly in the east.

In the preliminary studies conducted in 1971 —1972 on the occurrence of the *P. brassicae* races in Finland (LINNASALMI and PALONEN 1974), the races 2, 4, 6 and 7 were isolated. In two test places the races that were found in 1972 were, according to the present study, still the same, viz. race 4 in the test field of the Institute of Plant Pathology in Tikkurila (Vantaa) and race 7 in the test field of the Institute of Horticulture in Piikkiö. Ten years earlier, in 1964, samples from these localities were sent to Prof. P.H. WILLIAMS. He identified race 7 from Piikkiö and race 2 from Tikkurila (WILLIAMS 1966).

## Occurrence in different plant species

The races displayed the following distribution by different plant species: races 2, 3, 4, 6 and 7 were isolated from cabbage (60 isolates) and cauliflower (17 isolates); races 2, 3 and 7 from broccoli (4 isolates); race 4 from red cabbage and marrowstem kale (1 isolate from each); races 1, 2 and 4 from swede (5 isolates); and race 2 from turnip (2 isolates).

Of the cabbage cultivars grown in Finland, (cf. App. 2) only the Norwegian Resista and Respla (WEISAETH 1977) are partly resistant to some *P. brassicae* races. These cultivars were accepted for marketing in Norway in 1973 and in Finland in 1975. In our infection tests (glasshouse tests) with some of the *P. brassicae* races occurring in Finland, both cultivars were found to have some resistance to races 2, 4 and 7 when compared with the severely contaminated Blåtopp cultivar:

ra	ce (isol.)	cultivar, clubroot index (0-3					
_		Resista	Respla	Blåtopp (control)			
2	(F 118)	1.80	1.47	2.88			
3	(F 147)	2.36	2.49	2.96			
4	(F 155)	1.95	1.14	2.78			
7	(F 171)	1.86	0.84	2.52			

In some farms where these cultivars were grown in field sectors situated next to each other, severe contamination and consequently weaker growth of Blåtopp were evident, whereas Resista and Respla were only sparsely and slightly contaminated, and headed well.

## The occurrence of P. brassicae races determined according to Williams' system in other countries

#### Norway and Iceland

According to the working plan of NKJ project 27, the determination of *P. brassicae* races in Norwegian material was performed in Finland. The samples had mostly been taken from resistance breeding test fields in different parts of the country by the Norwegian project member, First Amanuensis G. WEISAETH. Half of the samples represented the breeding lines of cabbage. The test fields were located in 17 communes, and the number of samples totalled 38.

The race distribution was as follows:

race	communes no.	isolates no.	host plants
1	5	6	cabbage, swede
2	3	5	cabbage, swede, rape
4	13	21	cabbage, cauliflower, kohlrabi, swede
7	3	5	cabbage
9	1	1	swede

Detailed results from the race determinations of the Norwegian material, together with purified club material, were supplied annually to the Department of Vegetable Crops, Agricultural University of Norway, Ås, Norway. Some isolates from this material were used in our ECD test series (Table 3).

A report on the occurrence of races 1, 7 and 9 in Norway, in the Trøndelag region, is given in the publication of LINNASALMI and WEI-SAETH (1978). On the basis of our new sample material (cf. data given above), it would seem that race 4 is more common than the others elsewhere in Norway. There were some differences between the Finnish and Norwegian race spectra: race 9 was not found in Finland, and races 3 and 6, which are fairly common in Finland, were not found in the Norwegian material. However, data on races 3, 5 and 6, as well as races 1 and 9, are presented in some earlier studies on the Norwegian race spectrum (WEISAETH 1972). Two cabbage samples (WEI-SAETH'S breeding lines) from Iceland were analysed. Only race 7 was found in both samples (LINNASALMI and WEISAETH 1978).

#### Sweden

In connection with breeding work for clubroot resistance in cruciferous oil crops, R. JÖNSSON investigated the occurrence of *P. brassicae* races in Skåne, in southern Sweden. Using the method of WILLIAMS, JÖNSSON (1971, 1972) concluded that in the populations collected from various localities there was the possibility of occurrence of several *P. brassicae* races, and definite occurrence of race 15 in one case. Using the callus technique, JÖNSSON isolated races 1, 2, 3, 4, 6 and 7 (JÖNSSON 1981).

## Other countries

In addition to the Nordic countries, information about the occurrence of *P. brassicae* races has been published from the following countries: USA: races 6 and 7 (WILLIAMS and WALKER 1963, STRANDBERG and WILLIAMS 1967); Canada: races 1, 2, 3, 4, 6 and 6A (AYERS 1972), races 2 and 6 (CHIANG and CRÊTE 1972, REYES et al. 1974); Japan: race 2 (YOSHIKAWA and BUCZACKI 1978); GDR: races 1, 3, 4, 6, 7 and 9 (WILLIAMS and SEIDEL 1968); Poland: races 2, 4, 6 and 7 (Nowicki 1978); USSR: races 1, 6 and 7 (KRIVCHENKO et al. 1982). Moreover, mention should be made of the race identifications made by WILLIAMS (1966) on materials from the following countries: Czechoslovakia race 6; the UK race 1; FRG race 7; USSR race 2; Australia races 3, 6 and 7; New Zealand races 1, 2 and 4; Japan races 3 and 5.

# Comparison between the WILLIAMS' and ECD classification systems

In this study with the P. brassicae race ma-

Isolate		Race			s' system in diff. hosts		ECD code Diff. host groups
			JQ	BS	L	W	B.c. B.n. B.o.
N	P74048 C	1	22	0	83	86	16 — 31 — 12
F	117	2	97	92	100	0	16 - 03 - 31
F	118	2	90	93	97	0	16 - 19 - 31
F	1	2	79	83	97	0	16 - 19 - 31
F	147	3	96	1	99	0	16 - 18 - 30
F	155	4	89	68	94	80	16 - 31 - 31
F	168	4	76	60	82	74	17 - 31 - 31
F	106	6	68	0	0	0	16 - 00 - 30
F	171	7	94	86	0	0	16 - 00 - 31
Ν	P74046	7	94	88	0	0	16 - 02 - 31
I	P74040	7	86	70	0	0	16 - 02 - 31
I	P74043	7	91	69	0	0	16 - 03 - 31

Table 3. Comparison between the Williams' and ECD systems.

Diff. host symbols cf. App. 1. B.c. Brassica campestris; B.n. Brassica napus; B.o. Brassica cleracea; ECD codes according to Toxopeus 1974.

Origin of isolates from Finland (F) in App. 1; from Norway (N) P74048 C Jersey Queen (Asker) and P74046 Badger Shipper (Stjørdal); from Iceland (I) P74040 cabbage lines 666 Weisaeth and P74043 line 696 Weisaeth (Hruna-mannahreppur).

terial classified according to WILLIAMS' system (1966), a comparative test series was carried out using the ECD classification system (BUCZACKI et al. 1975). Pure isolates of races 1, 2, 3, 4, 6 and 7 (12 in total) were chosen for the tests. The results are shown in Table 3.

In the *Brassica campestris* group, the resistance and susceptibility of all differential hosts to the various races are the same, code number 16, with exception of one race 4 isolate, for which the code is 17 because of the susceptibility of differential host 01.

In the *Brassica napus* group, the differential hosts show more diversity in their reactions to the different races, but there is also variability with regard to isolates of the same race. For example, the codes for race 7 are 00, 02 and 03. Number 03 is also the code of one of the isolates of race 2. For the isolates of race 2, the reactions of the ECD differential hosts are to a large extent borderline cases between susceptibility and resistance, and therefore the code can be either 03 or 19. The differential hosts of the group do not serve to separate races 1 and 4. All are 100 % susceptible, code 31.

In the *Brassica oleracea* group, some differences were found. The code numbers are 12, 30 and 31. Code 12 applies only to race 1, but code number 30 to both races 3 and 6, and code 31 to races 2, 4 and 7.

Few results corresponding to those of our ECD test are found in publications from other countries. Code 16-31-12, which corresponds to race 1 in our material (isolate from Norwegian material), has been reported from France (ROUXEL et al. 1983). Code 16-31-31, corresponding to one of our race 4 isolates, has been reported from the USA (CAMPBELL et al. 1981) and from Scotland, UK (BROKEN-SHIRE and LEWIS 1981), and code 17-31-31, corresponding to the other race 4 isolate, also from the UK (BROKENSHIRE and LEWIS 1981, DIXON et al. 1981). The codes 16-02-31 and 16-03-31, corresponding to our race 7 (isolates from the Norwegian and Icelandic materials), were reported by DOBSON et al. (1983) from the USA. Data on code 16-02-31 have also been reported from Canada by CHIANG and CRÊTE (1983), who, however, mention it as the ECD code of their race 2 (sensu WIL-LIAMS). The survey of TOXOPEUS et al. (1986) of the ECD tests performed mostly in Western Europe up to 1982 includes code 16-31-12 (four occurrences), corresponding to our race 1; code 16-19-31 (two occurrences), corresponding to our race 2; codes 16-31-31 and 17-31-31 (65 and 5 occurrences respectively), corresponding to our race 4; codes 16-00-31, 16-02-31 and 16-03-31 (3, 51 and 13 occurrences respectively), corresponding to our race 7.

In addition to the studies mentioned above, determinations of *P. brassicae* pathotypes by the ECD system have been reported in the following publications: TOXOPEUS (1974) and TOXOPEUS and JANSEN (1975) from the Netherlands, HEYN (1981) from the FRG, JONES et al. (1982 a) from the UK, NAIKI et al. (1984) from Japan and LAMMERINK (1986) from New Zealand.

In comparing the advantages and disadvantages of WILLIAMS' method with the ECD method for isolating and classifying P. brassicae pathotypes, it can be concluded that WILLIAMS' system is a more time-saving method. By the cross-testing technique described previously (p. 418), it was possible to detect the main pathotypes in a large area (approx. 3 000 ha) by means of only four differential hosts. On the other hand, it is evident that with such a restricted set of differential hosts it is not possible to identify all possible pathotypes and pathotype variants with certainty. The ECD system, with its fifteen differential hosts, is much more laborious and the requirement for growing space is many times greater than with WILLIAMS' method. Even more serious is the fact that in our comparative test series, which was carried out with very clear and pure race isolates classified according to WIL-LIAMS' system, the ECD results showed uncertainties and contradictions, as can be seen in Table 3 and in the report of the results.

Soon after introduction of the ECD system, other workers also began to draw attention to

the uncertainties of the pathotype determinations obtained by the method. The studies of TINGGAL and WEBSTER (1981) showed that P. *brassicae* populations identified by the ECD method and assumed to be pure isolates in fact contained several pathotypes. Similarly, DIXON et al. (1981) and JONES et al. (1982 a, b) found mixtures of pathotypes in their collections. TOXOPEUS et al. (1986) mention uncertainties in coding *P. brassicae* populations by the ECD system. Worth mention is also the critical assessment of the limitations of the ECD method by CRUTE et al. (1980).

## Discussion

Regardless of the classification system employed, one of the most important requirements for the reliable determination of pathotypes is that a genetically uniform inoculum material can be obtained. One way to achieve this is the single spore technique starting from the resting spores of the P. brassicae fungus. Difficulties have been encountered in developing the technique. In the studies of BUCZACKI (1977), TINGGAL and WEBSTER (1981) and JONES et al. (1982 b), with ECD population materials, in which Brassica napus and B. campestris varieties were used as test plants, the results have not been very promising; the infection rate remained low, 20-30 % at best (TINGGAL and WEBSTER). Moreover, TINGGAL and WEBSTER found that when two single spore isolates from ECD populations were tested further, roughly one half gave a result that corresponded with the original code, whereas four new races differentiated from one of the populations and two new races from the other.

The single spore technique can obviously be improved, but judging by what is known thus far about the multistage internal life cycle of *P. brassicae* and its development in the host plant (INGRAM and TOMMERUP 1972, INGRAM 1978), and about the microstructure of the fungus as revealed by electron microscopy (DEKHUIJZEN 1979, IKEGAMI et al. 1978, BUC- ZACKI et al. 1979), it may be difficult to achieve a homozygotic fungus material. Theoretically there are many possibilities for different recombinations of differential pathogenicity genes (cf. CRUTE et al. 1980 with references, TINGGAL and WEBSTER 1981).

The significance of the genetic properties of the host plants used in the P. brassicae infection studies began to receive attention in the 1950s. MACFARLANE (1955) concluded that in certain cases the heterogeneity of the host plant population can cause variation in the infection results. In the 1960s WILLIAMS (1966), among others, stressed the importance of genetically uniform differential host materials in pathotype determinations. Since our preliminary work on P. brassicae races (LIN-NASALMI and PALONEN 1974), our aim has been to use homozygotic seed material of the differential hosts (cf. p. 417). This criterion was not always taken into account prior to the 1980s. Difficulties have been encountered in the production of homozygotic test plant lines by conventional methods. According to CRUTE et al. (1980), heterogeneity is apparent in some differential host lines of the B. oleracea group, possible in the B. campestris group and less likely only in the B. napus group, because the species is strongly inbreeding.

However, new developments have improved the prospects of producing homozygotic test plant material that has the appropriate resistance/susceptibility qualities. For instance, the production of haploid plants from anther cultures via embryogenesis will allow rapid establishment of pure lines. So far haploid plants have been obtained from various *Brassicaceae* species, e.g. *B. napus, B. campestris, B. oleracea* var. *italica* (KELLER and ARMSTRONG 1977, 1979, 1983), *B. oleracea* var. *gemmifera* (OCKENDON 1984) and *B. oleracea* var. *capitata* (CHIANG et al. 1985).

The rapid advance of plant molecular biochemistry and genetics offers new opportunities for studying questions of resistance to diseases. One of the new possibilities already in sight is the application of gene technology to modify the genom of a plant directly as desired. It remains to be seen how long it will take before the prerequisites exist for applying these techniques in the breeding of clubroot resistant cruciferous vegetable cultivars.

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Locality	Original ho	ost	(	Clubroo	ot index	1	Race
sample no.	species	cultivar	JQ	BS	L	W	×
Pohjois-Savo Sb							
193 Virtasalmi	swede	Mustiala	18	0	81	67	1
Varsinais-Suomi Ab							
97 Lohja	cauliflower	Erfurter	56	37	60	0	2
253 Masku	cauliflower		87	65	88	0	2
162 Raisio	cauliflower	Flora Blanca	93	94	96	0	2 2 2
169 Rusko	cabbage	Amager, hög	99	94	99	0	2
11 Rymättylä	cauliflower	Flora Blanca	47	68	78	0	
163 Sauvo	cauliflower	Flora Blanca	94	97	96	0	2
Uusimaa N							
1 Hyvinkää	cauliflower	Erfurter	79	83	97	0	2
76 Nurmijärvi	cabbage	Västernordland	93	87	95	0	2
86 Nurmijärvi	cabbage	Københavns Torve	90	57	77	0	2
Etelä-Karjala Ka							
154 Vehkalahti	cabbage	Ruhm von Enkhuizen	93	91	94	0	2
157 Vehkalahti	swede	Pandur	87	89	96	0	2
158 Anjalankoski	cabbage	Blåtopp Faales	94	94	99	0	2
Satakunta St							
196 Huittinen	cabbage	Futura	96	96	99	0	2
224 Kiikka	marrowstem kale		78	94	68	0	2
227 Lappi Tl	cauliflower		91	83	85	0	2
221 Mouhijärvi	broccoli	Greenia	53	71	41	0	2
Etelä-Häme Ta							
175 Asikkala	cabbage	Københavns Torve	85	70	87	0	2
176 Asikkala	cabbage	Blåtopp Faales	95	92	94	0	2
118 Hattula	cabbage	Ruhm von Enkhuizen	90	93	97	0	2
194 Kuhmoinen	turnip		93	69	96	0	2 2 2 2 2 2 2 2 2 2
205 Kuorevesi	turnip	Guldbåll	95	95	100	0	2
207 Lammi	cabbage	Blåtopp Faales	84	86	93	0	2
159 Orivesi	cabbage	Plåtopp Foolos	85 97	73 92	92 100	0	2
117 Sahalahti 199 Sahalahti	cabbage	Blåtopp Faales Blåtopp	94	89	99	0	2
209 Tuulos	cabbage	Diatopp	93	95	94	0	2
126 Tampere	cabbage		98	94	99	2	2
Etelä-Savo Sa							
143 Joutseno	cabbage	Golden Acre	97	29	86	0	2
	cabbage	Golden Acre	1	2)	00	0	2
Etelä-Pohjanmaa Oa							
233 Jalasjärvi	cauliflower		74	71	67	0	2
236 Vähäkyrö	cabbage		99	99	99	1	2
Pohjois-Häme Tb							
214 Toivakka	cabbage		95	56	99	0	2
Pohjois-Savo Sb							
192 Haukivuori	cabbage	Amager	81	68	94	0	2
Pohjois-Karjala Kb							
63 Eno	swede	Östgöta	70	49	74	0	2
182 Pyhäselkä	cabbage	Blåtopp Faales	93	75	95	0	2
183 Pyhäselkä	cabbage	Västernordland	86	57	94	0	2
177 Rääkkylä	cabbage	Københavns Torve	95	28	97	0	2 2 2
178 Rääkkylä	cabbage	Blåtopp Faales	98	68	94	0	2
179 Rääkkylä	cabbage	Ruhm von Enkhuizen	81	92	88	0	2
180 Rääkkylä	cabbage	Golden Acre	93	95	98	0	2

Differential plants: Jersey Queen JQ, Badger Shipper BS, Laurentian L, Wilhelmsburger W

<sup>1</sup> Each index is the mean of four replicates.

Locality	Original	host	(	Clubroo	t index	1	Race
sample no.	species	cultivar	JQ	BS	L	W	in south and the second
Keski-Pohjanmaa Om							
243 Kälviä	cabbage	Blåtopp Faales	86	78	89	0	2
Varsinais-Suomi Ab							
80 Vihti	cabbage	Blåtopp Faales	90	5	24	0	3
Uusimaa N							
70 Nurmijärvi	cabbage	Blåtopp Faales	88	6	90	0	3
Etelä-Karjala Ka							
149 Kymi	cabbage	Blåtopp Faales	95	9	93	0	3
Etelä-Savo Sa							
146 Imatra	cabbage	Golden Acre	72	1	66	0	3
147 Imatra	cabbage	Västernordland	96	1	99	0	3
142 Joutseno 144 Joutseno	broccoli cabbage	Greenia Ruhm von Enkhuizen	83 38	2	63 60	0	3
	cabbage	Kunm von Enknuizen	38	0	00	0	3
Pohjois-Savo Sb					0.2	0	
58 Karttula 197 Kuopio	cabbage	Plåtonn Foolos	84 91	6 4	93 97	0	3
139 Maaninka	cabbage cabbage	Blåtopp Faales	87	4	85	0	3
198 Suonenjoki	cabbage	Blåtopp Faales	92	11	67	0	3
Pohjois-Karjala Kb	ene enge	Diatopp Funite			0,	0	5
69 Joensuu	broccoli	Waltham	87	5	91	0	3
	broccon	Watthani	07	5	91	0	3
Varsinais-Suomi Ab	11.01						
228 Laitila 168 Turku	cauliflower cabbage	Amager hög	61 76	23 60	85 82	71 74	4
	cabbage	Amager nog	70	00	02	/4	4
Uusimaa N			10			60	
246 Tuusula 155 Vantaa	swede cabbage	Blåtopp Faales	49 89	34 68	82 94	69 80	4
	cabbage	Blatopp Faales	09	00	94	80	4
Satakunta St	11.01			12			
225 Eura 223 Kokemäki	cauliflower red cabbage	Flora Blanca	72 88	43 78	84 97	84 77	4
195 Köyliö	cauliflower	Igloo	81	54	98	97	4
229 Rauma	swede	19100	69	63	83	9	4
Etelä-Häme Ta							
220 Loppi	cabbage		85	58	95	81	4
Etelä-Pohjanmaa Oa	eucouge		00	50	,,,	01	
234 Ilmajoki	cauliflower		49	31	90	71	4
230 Isokyrö	cabbage	Marner Sepco	63	27	93	84	4
Pohjois-Häme Tb							
105 Jyväskylä	cabbage		82	30	87	85	4
Varsinais-Suomi Ab	eucouge		02	50	07	05	-
21 Sammatti	cabbage	Golden Acre	47	0	0	0	6
22 Sammatti	cabbage	Golden Acre	91	0 5	0	0	6
Etelä-Häme Ta	encouge			2	0	0	0
55 Hartola	cabbage	Igloo	26	0	0	0	6
122 Hauho	cabbage	Blåtopp Faales	61	2	1	0	6
112 Kangasala	cabbage	Blåtopp Faales	59	õ	0	0	6
124 Kangasala	cabbage	Blåtopp Faales	98	4	0	0	6
106 Pälkäne	cabbage	Västernordland	68	0	0	0	6
107 Pälkäne	cabbage		25	0	0	0	6
108 Pälkäne	cabbage	Blåtopp Faales	52	1	0	0	6
170 Toijala	cabbage		91	3	0	0	6

<sup>1</sup> Each index is the mean of four replicates.

Locality	Original	host	(	Clubroo	t index	1	Race
sample no.	species	cultivar	JQ	BS	L	W	
Pohjois-Savo Sb							
191 Haukivuori	cabbage	Blåtopp Faales	57	2	3	0	6
188 Joroinen	cauliflower	Erfurter	48	1	0	0	6
Pohjois-Pohjanmaa Ob							
67 Oulu	cauliflower		84	6	0	0	6
250 Oulu	cabbage		100	9	0	0	6
Varsinais-Suomi Ab							
171 Piikkiö	cabbage	TK 499 Weisaeth	94	86	0	0	7
Uusimaa N							
88 Espoo	cauliflower		97	19	0	0	7
Etelä-Häme Ta							
174 Hartola	cabbage	Toftegård	94	51	0	0	7
204 Längelmäki	cauliflower	Erfurter Dvärg	90	13	0	0	7
38 Kangasala	cabbage	Resista	100	27	0	0	7
Etelä-Savo Sa							
148 Lemi	cabbage	Blåtopp Faales	100	24	2	0	7
Etelä-Pohjanmaa Oa							
231 Alavus	cabbage		84	64	3	1	7
238 Laihia	cabbage	Futura	99	31	0	0	7
Pohjois-Häme Tb							
216 Jyväskylän mlk	broccoli		98	16	3	0	7
211 Keuruu	cabbage	Blåtopp Faales	100	21	0	0	7
Pohjois-Pohjanmaa Ob							
248 Oulu	cauliflower	Erfurter Dvärg	97	57	3	0	7

<sup>1</sup> Each index is the mean of four replicates.

## Appendix 2. The list of species and cultivars.

Scientific nomenclature according to Zander Handwörterbuch der Pflanzennamen (ENCKE et al. 1981).

	Brassica oleracea L.
Cabbage	convar. capitata (L.) Alef. var. capitata: Amager, Amager hög, Amager Stenhoved, Amager Toten, Blåtopp, Blåtopp Faales, Dala, Dima, Ditmarsker, Futura, Golden Acre, Håløyen Lundes, Københavns Torve, Marner Sepco, Resista, Respla, Ruhm von Enkhuizen, Septem- ber, Toftegård, Västernordland
Red cabbage	f. rubra: Amager Tagenshus, Haco
Cauliflower	convar. <i>botrytis</i> (L.) Alef. var. <i>botrytis</i> : Erfurter, Erfurter Dvärg, Flora Blanca, Grandessa, Igloo, Stor dansk
Broccoli	var. italica Plenck: Greenia, Waltham
Brussels sprouts	convar. <i>oleracea</i> var. <i>gemmifera</i> DC.: Jade convar. <i>acephala</i> (DC.) Alef.
Kohlrabi	var. gongylodes L.: Wiener glas
Curly kale	var. sabellica L.: cv. unknown
Marrowstem kale	var. viridis L.: cv. unknown
Chinese cabbage	Brassica pekinensis (Lour.) Rupr.: cv. unknown
	Brassica napus L.
Swede	var. napobrassica (L.) Rchb.: Messukylän lanttu, Mustiala, Pandur, Svensk gul, Wilhelmsburger, Östgöta
	Brassica campestris L.
Turnip	var. <i>rapifera</i> Metz. syn. <i>B. rapa</i> L. subsp. <i>rapa:</i> turnip Guldbåll, fodder turnip cv. unknown
	Raphanus sativus L.
Radish	var. radicula Pers.: cv. unknown

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## SELOSTUS

## Möhöjuuritaudin ja *Plasmodiophora* brassicae Wor. rotujen esiintyminen Suomessa

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Tutkimuksessa selvitettiin möhöjuuritaudin ja *Plasmodiophora brassicae* Wor. patotyyppien esiintymistä ristikukkaisissa (*Brassicaceae*) vihanneskasveissa Suomessa v. 1974—1978. Tarkastetuista 101 kunnasta 81 %:ssa esiintyi möhöjuurta. Tauti oli yleisintä maan etelä- ja keskiosissa, mutta sitä tavattiin myös ristikukkaiskasvien viljelyalueen pohjoisosissa (66—67° Pohj. lev.). Kertyneistä 375 kasvinäytteestä 65 % oli möhöjuurisia. Yleisimmin viljellyn kasvilajin, keräkaalin näytteistä (56 % aineistosta) 68 %, kukkakaalinäytteistä (22 % aineistosta) 63 %, muiden kaalilajien näytteistä (13 % aineistosta) 56 % ja ristikukkaisista juurikasnäytteistä (19 % aineistosta) 64 % oli möhöjuurisia. *P. brassicae* rotumääritykset tehtiin 90 näytteestä. Käytettiin Williamsin (1966) luokitussysteemiä. Eristetyt rodut olivat 1, 2, 3, 4, 6 ja 7. Rotu 2 oli ylivoimaisesti yleisin. Sitä tavattiin 32 kunnassa, rotuja 3, 4, 6 ja 7 kutakin 9—12 kunnassa, rotua 1 vain yhdessä kunnassa. Selviä eroja rotujen esiintymisessä maan eri osissa ei voitu todeta. Suoritettiin Williamsin ja ECD (Buczacki et al. 1975) luokitussysteemien vertailu. Williamsin systeemi todettiin nopeammaksi ja luotettavammaksi kuin ECD systeemi *P. brassicae* patotyyppien identifioimiseksi. Patotyypit määritettiin myös Norjassa ja Islannissa kerätystä möhöjuuriaineistosta.

Tutkimus on osa pohjoismaisesta möhöjuuriprojektista, NKJ-projekti 27 1974—1977: Möhöjuuren kestävyysjalostus, *Plasmodiophora brassicae* Wor. rodut sekä uusien torjunta-aineiden tehotutkimus.