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### Seria PHYTOPATHOLOGIA N. 75 — Sarja KASVITAUDIT n:o 75

# SEVENTY YEARS RESEARCH ON PLANT PATHOLOGY AND PLANT PROTECTION INSTITUTE OF PLANT PATHOLOGY 1911—1981

### AARRE YLIMÄKI

Ylimäki, A. 1981. Seventy years research on plant pathology and plant protection. Institute of Plant Pathology 1911—1981. Ann. Agric. Fenn. 20:61—73. (Agric. Res. Centre, Inst. Pl. Path. SF-01300 Vantaa 30, Finland).

During its early decades studies at the Institute of Plant Pathology were devoted to the systematics of fungi. The most extensive of the studies were the analyses dealing with smut fungi and Fusarium fungi. A study of diseases caused by a deficiency of boron was one of the first of its kind in the world. Objects of extensive studies in the 1940s and 1950s included overwintering of field plants, foot rot diseases of cereal plants, virus diseases of cereals and ley grasses, and storage diseases of potato, carrot and onion. The most important and extensive studies about cultivations under glass were those dealing with damping off and disinfection of the soil. In the 1960s studies were made of virus diseases of vegetables, potato, legumes and berry plants, and of the significance of fungus diseases of potato, clover, berry plants, vegetable plants and ornamental plants. Since the end of the 1960s increasing attention has been paid to the study of the quality of crops, the utilisation of the disease resistence of plants, the producing of healthy propagation material from potato and berry plants, and the investigation of harmful effects of pesticides. In addition to research, the Institute has throughout been responsible for implementing the plant quarantine prescribed by the Plant Protection Act and subsequently by the International Plant Protection Convention, and for the statutory evaluation of the efficiency and the registration of fungicides.

Index words: Plant diseases, plant protection, historical survey, research work, plant quarantine, testing of fungicides.

### ESTABLISHMENT AND SCOPE OF ACTIVITIES

The work of the Department for bacteriology, plant physiology and plant diseases at the State Agricultural-Economic Experimental Institute established by imperial decree in 1898 did not begin until June 1, 1911, at Tikkurila near Helsinki.

As systematic and biological studies of

fungi provide a foundation for research in fungus diseases of plants and their control, it was fortunate that J. I. Liro, an assistant at the University of Helsinki who from 1921 was extraordinary professor of plant biology and plant pathology, and who had already conducted extensive investigations of injuri-

ous fungi, became head of the department when it commenced. Thus the department and the instruction in agriculture at the University entered into a close inter-relationship.

Three periods can be distinguished in the work of the Department of Plant Pathology.

- 1. In the years 1911—1923 activities were much restricted on account of the small size of the monetary appropriations with the consequently small number of staff. In addition to the director, there was initially only a summertime assistant; and it was not until 1915 that the post of extraordinary assistant was established and until 1919 the post of a second assistant.
- 2. The activities of the department were substantially improved with the establishing of the Agricultural Experiment Institute in 1924 and with the acquiring of a position of its own in agricultural research by the Department of Plant Pathology, one of the nine departments of the Institute. The Plant Pro-

tection Law enacted at roughly that time gave the department additional tasks. Its research staff was consequently augmented, and from 1925 there were four assistant posts in addition to the director while from the beginning of the 1930s there were five assistant posts at the department. This was the situation throughout the 1930s. In the war years 1939—44 the activities of the department were almost entirely in abeyance on account of the staff's being engaged in military activities.

3. The period since World War II has been one of extremely vigorous development both in field cultivation and, especially, in garden cultivation. Efforts have been made to increase the amount of plant products by every means. Owing to the development of the chemical industry, new, efficient and easily used pesticides could be employed for plant protection.

In addition to research, the department was put in charge of other duties. Under provisions issued on the strength of the Plant

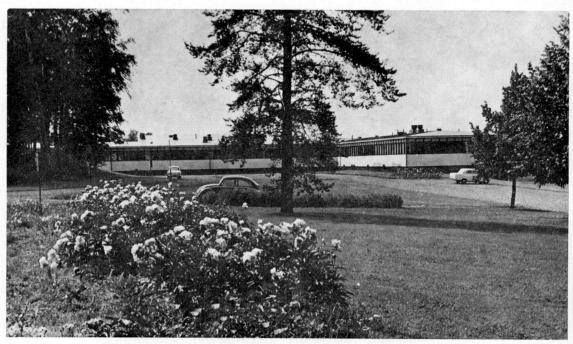


Fig. 1. A new laboratory building was completed in 1964.

Protection Act, supervision of the importing of living plants and plant parts (quarantine inspection) was improved. An extraordinary post of plant protection inspector had been set up at the department in 1943. The testing of the efficiency of plant protectants, and the supervising of their use and merchandizing, were commenced in cooperation with the Department of Pest Investigation under an Act of 1951.

From the beginning of 1957 the Agricultural Experiment Institute became the Agricul-

tural Research Centre, which has nine institutes of investigation, one of them being the Institute of Plant Pathology. The circumstances of the Institute improved substantially when a new and up-to-date laboratory building with pertaining greenhouses was completed in 1964.

The development of the Institute of Plant Pathology and its activities from 1911 to 1960 were described in detail when the Institute had been working for 50 years (Jamalainen 1961 a).

### RESPONSIBILITIES

The responsibilities of the Institute of Plant Pathology are to engage in research and testing in order to promote and develop agriculture, with the object of preventing damage caused by the diseases of cultivated plants. To that end the Institute

- investigates the distribution, damage and possibilities of controlling plant pathogens (bacteria, fungi and viruses). It also analyses physiogenic plant diseases caused by unfavourable growing conditions, and the means of preventing them
- engages in scientific and consultative publication

- endeavours to prevent the conveying of plant enemies to and from the country by controlling the state of health of imported and exported plants and plant products in accordance with regulations
- replies to inquiries concerning plant diseases and instructions for their control
- investigates the biological efficiency and usability of fungicides for the control of plant diseases before they are put on the market; participates in the registration and supervision of the import, manufacture, merchanizing and application of pesticides.

### STAFF

Directors (professors) of the Institute of Plant Pathology have been J. I. Liro 1911—37, A. J. Rainio 1937—43 (assistant 1919—37), E. A. Jamalainen 1944—69 (assistant 1925—44), A. Ylimäki 1970— (assistant 1946—47, plant protection inspector 1948—56, senior researcher 1957—70).

Assistants and researchers serving for lengthy periods have been A. Hilli 1925—34, H. E. Moliis 1925—35, V. B. Lehtola 1927 and

1933—45, H. Roivainen 1935—47, J. E. Hårdh 1945—55, Annikki Linnasalmi 1945—74, J. Mukula 1948—56, M. Haavisto 1949—58, Eeva Tapio 1954—73, K. Aura 1955—77, P. Talvia 1956—72, Katri Bremer 1957—64 and 1971—, Y. Rouvala 1962—73, E. Seppänen 1963—, A. Murtomaa 1965—72, Rauha Puttonen 1966—81.

Today, in addition to the post of director there are posts for one special researcher (Kaiho Mäkelä 1975—), two senior researchers (Katri Bremer 1971—, E. Seppänen 1963—) and three researchers (J. Kurtto 1973—, Kirsti Osara 1974—, R. Vanhanen 1974—) at the Institute.

The technical personnel include 14 regular employees as well as temporary research assistants and trainees depending on the available appropriations.

### CERTAIN EXTENSIVE SUBJECTS OF STUDY

A great deal of research into the systematics of fungi was performed at the Institute in its early decades. Major attention was devoted initially to the smut fungi of cereals and the diseases of potato but also to fungus diseases of sugarbeet, onion downy mildew and American gooseberry mildew. The efficacy of washing with formaline and of hot water treatments as devices of controlling smut fungi of seed grain was demonstrated, and these methods were introduced to some extent. Publication work from the period dealt with club root, ergot, stripe disease of barley, scab of apple and pear, and plum pockets. The report by Liro in 1923 on plant protection legislation abroad, and the appended proposal for the enactment of a plant protection law for Finland, turned out to be very important for the operations of the Institute and for the development of plant protection in Finland.

After Finland gained independence, farming and gardening began to acquire strength in every way. With it there was a growth in the understanding of the damage caused by plant diseases and the importance of controlling them.

### Cereal and ley studies

To control smut diseases and other seedborne fungus diseases, trials with the treatment of seed grain with mercury preparations were begun according to examples from abroad, and the dressing proved to be an excellent means in the control of seed-borne fungus diseases of cereals. The Department initiated an intensive information and counselling campaign to spread the use of the dressing method.

With the rapid expansion of the acreage cultivated with spring wheat, especially in western Finland and as far north as Central Ostrobothnia ever since the 1930s, and while there was specialisation in the one-sided cultivation of cereals on farms without cattle, difficulties began to occur in the growing of wheat, in particular, on account of the foot rot diseases. The most important pathogens of foot rot diseases are according to studies by Hårdh (1953) eyespot (Pseudocercosporella herpotrichoides) and by Ikäheimo (1959) takeall (Gaeumannomyces graminis), although some Fusarium species may also play an important part in the damage. According to extensive studies carried out in recent years, the Fusarium species and the Gaeumannomyces graminis are the most common causes of foot rot diseases, while eyespot is of lesser importance (Mäkelä and Parikka 1980).

Five virus diseases have been identified in studies done on spring cereals during the 1950s and 1960s: striate mosaic virus of wheat, and oat sterile-dwarf virus, barley yellow dwarf virus, Agropyron mosaic virus and brome grass mosaic virus. The oat sterile-dwarf virus caused great destruction in oats in areas verging on the Gulf of Bothnia in the 1950s. The studies indicated

that the disease can well be kept under control by using rye, wheat or two-row barley as nurse crop instead of oats (Bremer 1974).

As the weather prevailing during the grain harvest is usually quite damp in Finland, the crop is usually contaminated with fungi. As early as the 1930s Rainio found that the





Fig. 2. During the winters 1951—54 snow cover trials were carried out in order to investigate the effects of the snow cover on overwintering fields crops. Picture of the situation in the trial 1952/53, a. in March, b. in April.

grains of cereals and especially of oats are almost regularly heavily contaminated with Fusarium roseum (= F. graminearum) fungus and, as such, also poisonous to domestic animals, especially horses.

In the last decade the fungal flora was determined from a very large freshly threshed and partly also freshly stored cereal material, and it was found that it depends decisively on the treatment of the cereal after harvesting how much the fungi are able to lower the quality of the cereal. More than anything else, it is important how quickly the cereal can be dried. Particular attention has been paid to the capacity of some fungi to produce, as products of their metabolism, toxic or otherwise damaging agents, i.e. mycotoxins. The study has been pursued to some extent in cooperation with veterinarians and biochemists (Ylimäki 1970, Westermarck-Rosendahl and Ylimäki 1978, Korpinen and Ylimäki 1972 a and b, Korpinen, Kallela and Ylimäki 1972, Ylimäki et al. 1979).

### Studies on overwintering

In the latter half of the 1940s right after the war years, the causes of poor keeping through the winter of overwintering cereal plants was made a subject of investigation, and this work went on into the 1960s. In respect of leys in northern Finland, the study has been taken up again in recent years.

In perennial studies on the significance of the snow cover, it was shown that in the region of heavy snow cover, i.e. in the central, eastern and northern parts of the country, plants are well protected in winter from the ravages of ground frost or of needle ice in the soil (Ylimäki 1962). Under the snow, they are vulnerable to destruction from winter-killing fungi, especially if the snow

in autumn falls on unfrozen ground. But in the coastal areas of southern and western Finland where snow is scanty, the abiotic severity factors of winter, i.e. stagnant water, covering ice, ground frost or needle ice in the soil and freezing temperatures, cause destruction to plants almost every year instead.

The winter-killing fungi that is most common and most destructive to winter cereals and ley grasses is snow mould (Fusarium nivale) while in many winters particular destruction has been caused also by Typhula fungi (T. ishikariensis and T. incarnata) in the central, eastern and northern parts of the country and by Sclerotinia borealis in North Finland (Jamalainen 1956).

In respect of clover rot (Sclerotinia trifoliorum) which is generally destructive to ley legumes especially red clover, improved clover strains and local clover strains show considerable variations in resistance although all clovers may suffer serious damage in autumn and winter, which are extremely favourable to the disease (Ylimäki 1969).

In the 1950s and 1960s the Institute of Plant Pathology performed extensive resistance tests with destructive winter-killing fungi on improved strains of winter cereals and of ley grasses despatched for testing from plant breeding stations in other Scandinavian countries. These tests performed in the laboratory and field, show that there were such great differences in the resistances of various plant species and varieties to destructive winter-killing fungi, that it was found possible to increase the resistance to destructive winter-killing fungi through plant breeding (Jamalainen 1969, 1974).

In connection with studies on the growth requirements of clover and, in particular, its overwintering, attention had been paid from as early as the 1950s to root browning and decay of clover even when clover rot did not occur. Seedlings in leys were found to die with symptoms of damping off. Certain fungi were regularly found in the decaying roots, i.e. mainly species of Fusarium, Cylindrocarpon and Rhizoctonia. These common soil fungi were found to penetrate into the roots of clover, especially through wounds. The root decay has been found to be even more damaging than clover rot in leys, because it causes destruction all the time and not merely in occasional years as does clover rot (Ylimäki 1967).

Since the latter half of the 1940s the Institute has performed quite extensive treatment tests on shoots of winter rye and winter wheat and clover stands, with chemicals for

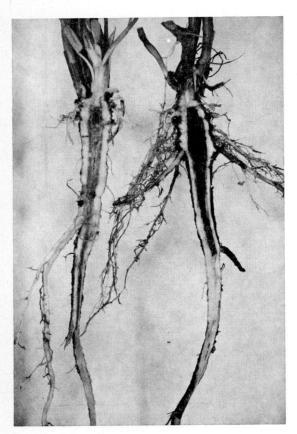


Fig. 3. Clover root rot.

the control of destructive winter-killing fungi. Among the numerous tested chemicals, treatment of the stands in autumn with quintozene, i.e. PCNB preparations, proved to be extremely effective in the control of destructive winter-killing fungi (Jamalainen and Ylimäki 1956, Ylimäki 1969). It has subsequently been carried out in practical farming, chiefly on fields of winter cereal and cultivations of red clover seeds. It has been found in recent years that benomyl and thiophanatemethyl preparations, too, are effective against snow mould (Fusarium nivale) of winter cereals and ley grasses.

### Potato

The occurrence of potato late blight (Phytopthora infestans) and its destruction vary a great deal in Finland from year to year. According to studies made by Seppänen (1971), leaf blight occurs in the south of Finland almost every year, however, and on average every third year there is a severe blight year. In the worst years the reduction of the potato crop has been 40 per cent, though the average is under 10 per cent. In Ostrobothnia and in North Finland the impact of the blight is less, although it can be extremely damaging there some years too, especially on clay soil.

Previously tuber blight was regarded as being the most serious storage disease of potato. With mechanisation of potato cultivation, the importance of wound parasites of tubers has increased. Studies in recent years have shown that fungi of the genera Fusarium and Phoma, in particular, cause considerable storage losses (Seppänen 1972 a, 1980).

A fairly large survey of the external quality of food potato in commerce, and factors

affecting it, was performed in 1967—70 (Seppänen 1972 b).

Potato ring rot (Corynebacterium sepedonicum) was found in Finland for the first time in 1970. Studies revealed that it had spread the most in Ostrobothnia but it was also found locally elsewhere in the country (Seppänen and Heinämies 1972).

Virus diseases of potato are calculated to have caused an annual loss of about 20 million marks in Finland during the 1970s. Most common were the X- and S-viruses, but with specialisation in the cultivation of potato the importance of the insect-borne A-, M- and Y-viruses has increased (Seppänen 1972).

After the studies begun in the early 1970s aiming at the production of seed potatoes free from virus (Tapio 1972), it was thought there was reason and opportunity to set up a Seed Potato Centre at the Agricultural Research Centre in 1976 to carry on production of virus free potatoes in Finland.

### Vegetables

At the turn of the 1940s—1950s damping off fungi occurring in cultivations under glass, and the control of them, were the object of thorough studies. Soil disinfection tests associated with these studies laid a foundation for the employment of the methods of soil steaming and chemical disinfection on cultivations under glass in Finland (Linnasalmi 1952). In experiments on the control of club root (*Plasmodiophora brassicae*) performed at roughly the same time mercurous chloride preparations proved to be effective both on swede and cabbages.

The resistance of foreign swede and white cabbage to club root has been subject to testing since the 1950s, and oil plants were included later in the tests. In the 1970s the investigations expanded into inter-Nordic

NKJ (The Scandinavian Contact Agency for Agricultural Research) projects in which the occurrence of strains of club root in Finland was investigated under professor Linnasalmi, and the resistance of improved white cabbage plants to club root. As a result of this cooperation, white cabbage cultivars resistant to club root were brought on the market.

Tobacco mosaic virus (TMV) caused substantial destruction in greenhouse tomatoes. Once the TMV types occurring in Finland had been established, it was found in resistance studies that foreign cultivars bred for resistance to TMV are also resistant to the Finnish TMV types (Murtomaa 1966, Linnasalmi 1980).

In studies of virus diseases of cucumber the occurrence and importance of these diseases and the characteristics and chemical composition of the viruses were studied (Linnasalmi 1966, Linnasalmi and Toiviainen 1974, 1975).

As grey mould of onion had caused great damage in stored multiplier onion in North Finland in the 1940s and 1950s, extensive studies were conducted on the susceptibility of Finnish onion material to this disease, and on the effect of the predrying of onions on the storage. The effects of heat treatment on the emergence of inflorescences, on flowering and on the yield of multiplier onion were also studied (Aura 1963, 1968). It was found that a virus disease was the cause of the degeneration of multiplier onion. As onion sets were not found to be suceptible to the virus disease, these were taken into use particularly in the south and central parts of the country.

In 1974 white rot of onion (Sclerotium cepivorum) was found to have caused considerable damage to onion cultivations in Åland and certain areas of mainland Finland. Investigation of the spread and control of this

disease was incorporated into the program of the Institute.

In her study extending to all the Nordic countries, Tapio (1970) isolated bean yellow mosaic virus and pea mosaic virus from several species of legume. The impact of these viruses in Finland was found to be of minor importance.

An extensive study of the storage of carrots was performed by Mukula (1957), in which 16 species of fungi were specified as being destructive to carrot. Storage tests revealed that technazene preparations were effective against *Sclerotinia sclerotiorum* and *Botrytis cinerea* fungi, which are among the most damaging of pathogens.

In very recent years investigations have been made of the effect of strains of downy mildew of lettuce (*Bremia lactucae*) (Osara 1978), of root diseases of cucumber and of wilt disease of tomato.

### Fruit and berry plants

In the control of apple scab, the spray timing method was found in the 1950s to be effective in Finnish circumstances (Hårdh 1956), and it was generally adopted by commercial apple growers.

Since the 1950s the growing of berries has become a considerable source of income in many parts of Finland, and cultivation has undergone expansion. With the intensification of cultivation, increasing attention has been paid, where pests and diseases are concerned, especially to grey mould of strawberry and gooseberry mildew. Root rot was found to be damaging to cultivations of strawberry that had taken the form of commercial monoculture (Ylimäki 1970). The department began to produce virus-free propagation material of raspberry, strawberry, currents and of a hybrid Rubus idaeus  $\times R$ . arcticus, and this experimental phase led to the establishment of a Healthy plant propagation farm in 1976, this new unit being responsible for the production of virus-free mother plants approved for the purpose (Bremer and Ylimäki 1978). This is being done with the aid of the Institute of Plant Pathology and the Institute of Pest Investigation.

### Other studies

Most among the mycological studies have been the studies by Liro of smut fungi. From the very abundant material collected by him, he determined several entirely new species of smut fungi (Liro 1924, 1938). He, together with his colleagues, collected samples of other fungi also, and as a result of this work the mycological collection *Mycotheca Fennica* was published, which comprises a total of 900 species of fungi. A summary of *Fusarium* fungi encountered in Finland was made by Jamalainen (1970).

The boron deficiency diseases were investigated in the 1930s in Finland by Jamalainen, who showed that brownheart disease of swede was due to a deficiency of boron. Likewise, internal cork disease of apples was found to be caused by a deficiency of this element. In sugar beet cultivations a common disease especially on heavily limed fields was heart rot of sugar beets. Trials carried out showed that these diseases could be prevented by applications of boron (Jamalainen 1949).

With the very marked increase in the production of ornamental plants from the 1950s, increasing trouble was experienced from such diseases as carnation wilt, many diseases of flower bulbs, powdery mildew of rose and of begonia, etc. The studies concerning these have led to better hygiene in greenhouses, to disinfection of soil to effective attention to certain technical factors of cultivation and to the regular employment of many control functions such as the control of powdery mildew fungi.

Forest plantation derived considerable benefit from the activities of the Institute through the studies on control that were conducted in the 1950s in forest nurseries on needle cast of pine (Lophodermium pinastri), and black snow mould (Herpotrichia nigra)

and pine seedlings snow blight (*Phacidium infestans*) which had until then regularly destroyed spruce seedlings. As a result, the control of these diseases was adopted as a regular step in cultivation at nurseries (Jamalainen 1961 b).

### COOPERATION WITH OTHER RESEARCH INSTITUTIONS

Owing to the relatively limited researcher resources, and also for reasons of expediency, the Institute of Plant Pathology has consistently cooperated with other institutes of the Agricultural Research Centre (Agricultural Experiment Institute) and, especially, with the experimental stations and also with outside research institutes. Cooperation has been particularly useful with some of the com-

mercial cooperatives, with the foodstuffs industry with field and garden producers and with some of the consultative organisations of these. From the start, the researchers of the Institute have taken part in the instruction at the University of Helsinki in the capacity of docents and as course and seminar lecturers.

### INTERNATIONAL ACTIVITIES

The research work requires keeping up constantly with research work being done abroad. The Institute has also endeavoured to obtain for its library the most important journals and manuals, while researchers have attended the most important international congresses, seminars and other such meetings, where ever possible. Nordic cooperation has traditionally been prominent,

but in recent years participation has been extended to other parts of Europe too. Some researchers have had the opportunity to acquire additional education at universities or research institutions abroad, and the Institute has similarly received visiting researchers from other countries. Researchers have participated in the activities of the numerous international organisations.

### STATUTORY PLANT PROTECTION ASSIGNMENTS

In accordance with the Plant Protection Act of 1925 and the commitments to subsequent international treaties, the Institute of Plant Pathology together with the Institute of Pest Investigation has acted to prevent the entry to, and spreading in the country of »dangerous plant destructors».

When potato wart disease had been encountered in Finland for the first time, in autumn 1924, energetic preventive measures were initiated by Liro against the spreading of this disease by means of quarantine and cultivation restriction allowed by the Plant Protection Act. Dozens of wart-resistant varieties of potato were obtained from abroad for testing in Finland. Owing to these steps, which were subsequently maintained, the spreading of potato wart disease has been

successfully curbed in Finland. When the post of plant protection inspector was established at the Institute in 1943, statutory plant protection and import and export inspections of plants could be conducted in 1947. Since 1974 responsibilities devolving from the Plant Protection Act have been discharged by the plant quarantine unit, a joint organ of the Institute of Plant Pathology and the Institute of Pest Investigation, which has offices in Helsinki and Turku and local inspectors in other localities. Under the new Plant Pro-

tection Act issued at the beginning of this year, these responsibilities have been transferred in their entirety to the National Board of Agriculture as of March 1, 1981.

Since 1952 the Institute of Plant Pathology has carried out statutory testing of the biological efficacy and the usability of fungicides, and together with the Institute of Pest Investigation and subsequently with the Institutes of Plant Husbandry and Horticulture, under the Pesticides Act of 1969, has been responsible for registration of pesticides.

### INFORMATION AND COUNSELLING

The results of the studies have been made available for use by researchers, counsellors and growers in the form of articles published in domestic and foreign series of scientific publications, journals and guidance booklets. The scientific articles were previously published in the form of Publications of the Finnish State Agricultural Research Board or the Journal of the Scientific Agricultural Society of Finland or Acta Agralia Fennica. Since 1962 the scientific papers have mainly appeared in the Annales Agriculturae Fenniae, the Agricultural Research Centre's own journal.

Ever since the foundation of the Institute, its researchers have been in close contact with farmers and gardeners and have supplied them with instructions for the control of plant diseases. Members of the Institute have also participated in education and counselling as lecturers.

The intensification and expansion of garden and orchard cultivation and the expansion of chemical protection of plants were the reasons that the post of plant protection advisor was established jointly at the departments of Plant Pathology and of Pest Investigation during the 1950s, with the contribution of the horticultural organizations. Primary duty of the holder of this post is to maintain contacts with counselling organisations, to organise plant protection courses and to give lectures. He has also been much occupied the whole time with direct counselling service for growers.

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### **SELOSTUS**

### Seitsemän vuosikymmentä kasvitauti- ja kasvinsuojelututkimusta Kasvitautien tutkimuslaitos 1911—1981

### AARRE YLIMÄKI

### Maatalouden tutkimuskeskus

Toiminnan ensimmäisinä vuosikymmeninä suoritettiin kasvitautien tutkimuslaitoksessa pääasiassa sienisystemaattisia tutkimuksia, joista merkittävimpiä olivat viljojen nokisieniä sekä Fusariumsieniä koskeneet selvitykset. Boorin puutoksesta aiheutuvista taudeista suoritettu tutkimus oli yksi ensimmäisiä maailmassa.

1940- ja 1950-luvuilla olivat laajimpien tutkimusten kohteina peltokasvien talvehtiminen, viljojen tyvitaudit, viljojen ja nurmiheinien virustaudit sekä perunan, porkkanan ja sipulin varastotaudit. Lasinalaisviljelyksillä suoritettiin laajoja ja merkittäviksi muodostuneita taimipoltetta ja mullan desinfiointia koskeneita tutkimuksia.

1960-luvulla selvitettiin sieni- ja virustautien merkitystä vihanneskasveissa, perunassa, palkokasveissa ja marjakasveissa. Vuosikymmenen puolivälistä alkaen on kiinnitetty yhä enenevässä määrin huomiota satojen laatuun vaikuttavien tekijöiden tutkimiseen, kasvien taudinkestävyyden hyväksi käyttämiseen, terveen lisäysaineiston tuottamiseen perunasta ja marjakasveista sekä torjunta-aineiden haittavaikutusten selvittämiseen.

Tutkimustyön lisäksi on suoritettu monimuotoista kasvinsuojeluneuvontaa, huolehdittu kasvinsuojelulain ja myöhemmin myös kansainvälisen kasvinsuojeluyleissopimuksen edellyttämästä kasvintarkastustoiminnasta sekä niinikään lakisäteisestä kasvitautien torjunta-aineiden tehokkuuden tarkastamisesta ja valmisteiden rekisteröinnistä.

### Seria PHYTOPATHOLOGIA N. 76 — Sarja KASVITAUDIT n:o 76

### THE MYCOFLORA OF CEREAL SEEDS AND SOME FEEDSTUFFS

### AARRE YLIMÄKI

Ylimäki, A. 1981. **The mycoflora of cereal seeds and some feedstuffs.** Ann. Agric. Fenn. 20: 74—88 (Agric. Res. Centre, Inst. Pl. Path., SF-01300 Vantaa 30, Finland).

A survey was carried out in 1966-1973 in order to obtain a general picture of the microbial population on cereal grain at harvest time. The microflora of 2601 seed samples of rye, winter wheat, spring wheat, barley and oats originating from different parts of Finland were investigated. Alternaria, Cladosporium, Fusarium and Penicillium were the most frequent fungus genera on seed samples (on 90-100 %, 73-100  $^{0}/_{0}$ , 56—100  $^{0}/_{0}$  and 47—100  $^{0}/_{0}$  of samples, respectively). Fusarium culmorum (W. G. Smith) Sacc., F. poae (Pk.) Wr., F. tricinctum (Cda.) Sacc., F. avenaceum (Fr.) Sacc., F. arthrosporioides Sherb, F. graminearum Schwabe and F. oxysporum Schl. emend. Snyd. & Hans. were the most common Fusarium species. F. nivale (Fr.) Ces. was a curiosity, cccurring only very rarely. In 467 feedstuff samples, composed of feed grains, commercial feed mixes, hay and silage, Penicillium, Mucor and Rhizopus fungi were by far the most frequent, but the bacterial contamination was also very heavy. A total of 113 fungi belonging to 62 genera were identified in this survey.

Index words: Cereals, feedstuffs, microflora, mycoflora, fungi, snow mould, Alternaria, Cladosporium, Fusarium arthrosporioides, F. avenaceum, F. graminearum, F. nivale, F. poae, F. tricinctum, Mucor, Penicillium, Rhizopus.

### INTRODUCTION

In Finland, as elsewhere in Scandinavia at similar latitudes, the growing season is short and the harvesting time usually much wetter than the early part of the growing season. Rain showers make combine harvesting difficult and great quantities of cereals often become mouldy. Lodging and germination in the ear on the field greatly lower the quality of the grain, and under these conditions many micro-organisms regularly contribute to a deterioration of the situation.

Even in the most favourable years the moisture content of grain is about 25 %, but during rainy harvest times it can be 40 per cent. To prevent sprouting in the ear, harvesting is started immediately after the moisture content has fallen to about 30—35 %.

It is clear that when combine harvested, such moist grain is particularly susceptible to spontaneous heating and other factors which diminish its quality (Westermarck-

Table 1. The quality of cereal yields in 1966—1975 according to Monthly Reviews of Agricultural Statistics (Anon. 1966—1976).

		Total	yield milj	. kg		Qualitat	ively sale	eable y	ield in per	r cents
Year	Winter wheat	Spring wheat	Rye	Barley	Oats	Winter wheat	Spring wheat	Rye	Barley	Oats
1966	67,2	301,1	118,6	596,7	880,8	90	85	87	86	78
1967	161,1	345,7	162,7	680,8	939,9	56	65	59	79	82
1968	160,0	355,5	133,9	773,9	1063,7	92	83	81	75	81
1969	212,8	268,6	125,8	840,0	1137,7	95	91	90	87	89
1970	146,4	262,9	131,4	933,4	1329,7	90	87	81	76	81
1971	137,1	306,3	131,8	1054,2	1423,7	94	92	91	83	84
1972	142,8	319,8	118,6	1140,2	1245,3	95	77	91	81	81
1973	157,0	305,0	124,2	992,4	1169,4	96	93	95	85	83
1974	134,5	458,6	134,4	962,9	1112,8	86	54	79	74	69
1975	129,5	492,0	80,7	1241,9	1450,1	97	95	95	88	89

Rosendahl and Ylimäki 1978, Table 1) and it is also an excellent growing medium for several micro-organisms. As a general rule, grain that is unsuitable for seed or bread is used as feed for domestic animals.

Since the beginning of the 1960s, the Institute of Plant Pathology has, each year, received mouldy grain samples from different sources, with requests to investigate their suitability for bread or for feed. In some cases veterinary surgeons had suspected that illness in animals had been caused by the contaminated grain.

There is certain evidence concerning the

ability of some fungi to synthesize highly toxic compounds, mycotoxins, which on consumption can cause very serious diseases in both man and animals (Forgacs and Carll 1962, Brook and White 1966). Knowledge of the microflora of cereal grains, and of the different feedstuffs in Finland was, however, very superficial. A study was therefore carried out in 1966—1973 in order to obtain a general picture of the microbial population on cereal grain at the time of harvesting. In addition to this, a number of samples of various feedstuffs e.g. feed grains, commercial feed mixes, hay and silage were investigated.

### **MATERIALS**

### Cereal grains

A total of 2601 grain samples were collected from different parts of the country during harvest time in 1966—1973 (Table 2). The majority of these samples were received from the various research units of the Agricultural Research Centre (Fig. 1). In addition, a number of samples were received from the State Granary, Helsinki, Work Efficiency Association, Helsinki, grain dealers and private

farms in different parts of the country (Table 3).

Most of the samples were combine harvested and dried immediately or soon afterwards in warm-air grain dryers. However, some of the samples were dried in cool-air dryers. After drying to about 15 per cent moisture, the seed samples were sent to the Institute of Plant Pathology for examination. Samples were kept in paper bags until examination. In general, the samples were examined im-

Table 2. The number of seed samples examined in 1966—1973.

		Num	ber of sa	amples		
Year	Rye	Winter wheat	Spring wheat	Barley	Oats	Total
1966	_	_	66	60	_	126
1967	110	288	405	104	_	907
1968	57	68	237	76	24	462
1969	10	11	50	15	_	86
1970 - 71	6	_	1	32	11	50
1972	12	2	201	349	155	719
1973	17	20	46	96	72	251
Total	212	389	1006	732	262	2601

mediately but some samples had to wait 2—4 weeks.

### **Feedstuffs**

The majority of all 467 feed samples originated from the various research units of the Agricultural Research Centre, but numerous samples were also received from private farms, veterinary surgeons and commercial stores (Table 3).

A number of feed samples, especially those sent by veterinary surgeons, originated from cases when these feeds or fodders were suspected as the cause of a single case of disease or an outbreak of toxicosis among farm animals.

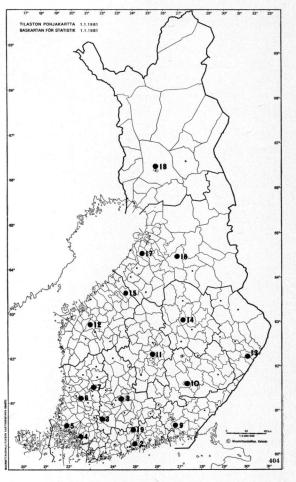


Fig. 1. Institutes and Experimental Stations of the Agricultural Research Centre (cf. Table 3).

### **METHODS**

The cereal seeds and the feeds were examined using a somewhat modified version of the blotter test (de Tempe 1963). Seeds were placed in Petri dishes (Ø 14 cm) or in germination bowls (Ø 20 cm), 25 seeds per dish. There was a layer of cellulose wadding and filter paper, both saturated with sterilized water, on the bottom of the dishes or bowls.

At the start of the investigation, the grains were surface sterilized with 0,3 per cent oxykinolinesulphate and alcohol, whereafter they were washed with abundant sterilized water. This procedure was later abandoned, since the fast growing saprophytes did not seem to inhibit other harmful fungi when growing on plain filter paper.

The bowls were first kept in a cold

Table 3. Origin of samples 1966-1973.

Origin	No. in Fig. 1	Cereal grains	Feed stuffs
Agricultural Research Centre			•
Institute of Animal Husbandry, Tikkurila	${f 2}$	X	×
Institute of Plant Husbandry, Tikkurila	2	X	
Institute of Plant Breeding, Jokioinen	3	X	
South West Finland Experimental Station, Mictoinen	5	×	×
Satakunta Experimental Station, Kokemäki	6	×	
Sata-Häme Experimental Station, Mouhijärvi	7	X	
Häme Experimental Station, Pälkäne	8	×	
Kymenlaakso Experimental Station, Anjalankoski	9	×	
South Savo Experimental Station, Mikkeli	10	×	
Central Finland Experimental Station, Laukaa	11	×	×
South Pohjanmaa Experimental Station, Ylistaro	12	X	
North Savo Experimental Station, Maaninka	14	×	×
Kainuu Experimental Station, Vaala	16	×	
North Pohjanmaa Experimental Station, Ruukki	17	×	×
Lapland Experimental Station, Rovaniemi	18	, ×	×
Swine Research Station, Hyvinkää	19		X
State Granary		×	×
Milling industry		×	
Grain dealers		X	
Work Efficiency Association		×	
Private farms		×	×
Veterinarians		×	×

chamber (5—8°C) for 3—5 days, and then subjected to UV light for two hours before being placed in the laboratory at a temperature of 20—22°C for 18—21 days.

After the 14 day incubation period, the germination rate of the seeds was counted and the sprouts were cut to facilitate microscope examination. The microflora was premilinarily determined with a stereomicroscope. For more accurate species determination of the fungi, hyphal tip and single spore isolates were taken and grown on oat meal agar (pH 4,4—5,0) or potato dextrose agar (PDA) (Difco 0013—01) to observe the speed of growth, colour and other colony characteristics. For more precise species determination, compound microscope slides were pre-

pared using the lactophenol technique or distilled water to which a drop of safranine was added (Fusarium).

The identification of the fungi was principally based on spore characters. The three week incubation period was found to be necessary for good sporulation of some species, mainly those of *Epicoccum* and *Fusarium*.

In the classification of the fungi, the system of Ainsworth (1971) has been followed. The nomenclature of Fusarium used in this work is based on that of Gordon (1952, 1960) except for the Fusarium section Sporotrichiella which is classified according to Seemüller (1968). Other Deuteromycotina were identified according to Barron (1968), von Arx (1970) and Ellis (1971, 1976).

### RESULTS

1. The composition of the microflora of cereal seeds.

Tables 4 and 5 give the frequency of the different micro-organisms found in 1966—



Fig. 2. Barley ears contaminated by Fusarium.

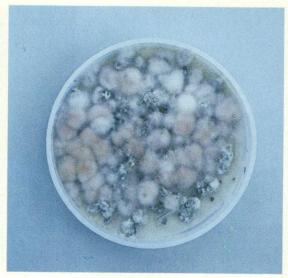


Fig. 3. Feed grain contaminated by Fusarium.

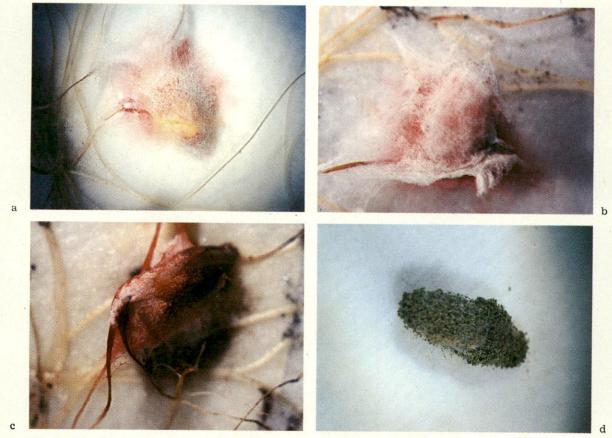


Fig. 4. Colonies growing from contaminated grains, a. Fusarium culmorum, b. F. avenaceum, c. F. tricinctum, d. Aspergillus flavus.

Table 4. The frequence of various fungus genera and bacteria in cereal seed samples in 1966—1971.

<u> </u>					
•	$\mathbf{R}\mathbf{y}\mathbf{e}$	Winter wheat	Spring wheat	Barley	Oats
	%	%	%	<b>%</b>	%
Number of samples	183	367	759	287	35
ZYGOMYCOTINA					
Mucor	22,5	6,2	12,1	8,5	6,8
Mycotypha	3,8	2,6	1,9	0	7,6
Rhizopus	22,2	18,7	15,9	18,5	8,7
ASCOMYCOTINA	•	•		•	
Chaetomium	1,5	1,6	3,0	4,7	1,8
Sordaria	0	0	0,5	0,3	0
DEUTEROMYCOTINA					
Hyphomycetes, light					
Acremoniella	2,1	1,1	. 0,6	2,3	27,4
Arthrobotrys	0,9	3,7	4,8	1,5	1,1
Cephalosporium	18,5	12,3	10,8	36,7	82,1
Fusarium	67,1	56,0	61,0	82,0	100
Fusarium Gliocladium	3,0	5,0	1,6	4,1	0
Giiociaarum Gonatobotrys	2,7	/ 0,5	9,7	1,7	0
	0	0	0,4	0,4	0
Oedocephalum	3,5	1,4	3,9	1,4	12,3
Papulaspora	86,0	83,0	67,8	62,8	47,1
Penicillium	0,3	1,0	0,3	2,0	3,2
Rhizoctonia	2,9	0,5	0,5	6,6	, O
Torula	2,1	1,5	0,5	1,4	2,9
Trichoderma	5,4	2,9	10,8	2,4	9,2
Trichothecium	2,3	0,5	0,3	0,3	2,5
Verticillium	2,0	0,0	0,0	0,0	_,-
Hyphomycetes, dark	91,1	97,9	94,3	98,7	100
Alternaria	19,0	6,7	10,6	3,1	0
Arthrinium		2,9	4,9	4,5	3,6
Aspergillus	1,5	7,4	11,9	10,6	48,3
Botryotrichum	12,1	9,3	6,4	3,4	0
Botrytis	11,7	0,5	0,4	0,4	0
Chlamydomyces	0,6	91.3	72,9	74,3	100
Cladosporium	93,8		12,5	0,2	0
Doratomyces	0	0,5	1,1	9,9	6,7
Drechslera	0,3	0,4		57,0	62,8
Epicoccum	45,2	57,2	57,8	10,1	2,7
Monodictys	9,7	6,5	4,1	10,1	2,1
Stachybotrys	0,6	1,0	0,1	•	18,5
Stemphylium	41,8	25,3	22,3	13,1 18,6	10,0
Trichocladium	19,3	15,1	8,8	10,0	U
Coelomycetes, Melanchoniales		0.5	0.0	e 9	6,2
Colletotrichum	1,2	0,5	8,3	6,3	0,2
Coelomycetes, Sphaeropsidales	_	•		กว	C
Septoria	0	0	2,0	2,3 17.2	(
OTHER FUNGI	6,9	21,8	27,6	17,3	64,3
BACTERIA (Streptomyces)	51,0	61,2	55,7	75,7	04,0

1971 and 1972—1973 in seed samples, and Tabel 6 gives their frequencies in single seeds.

Alternaria was by far the most frequent fungus: it occurred in more than  $90\,^{0}/_{0}$  of all samples of various cereals. The following most frequent fungus genera were Cla-

dosporium, Fusarium and Penicillium, occurring in 73—100 %, 56—100 % and 47—100 % of samples, respectively. Oat samples in particular showed very high contamination by Alternaria, Cladosporium and Fusarium fungi in all these years.

The contamination of single seeds was

Table 5. The frequence of various fungus genera and bacteria in cereal seed samples in 1972—1973.

	Rye	Winter wheat	Spring wheat	Barley	Oats
		<u></u> %	%	<b>%</b>	%
Number of samples	29	21	235	410	207
MYXOMYCOTA					
Myxomycetes					
Physarum	0	0	0	6,3	9,7
ZYGOMYCOTINA		-	·	0,0	٥,١
Mucor	69,0	71,4	66,0	62,9	44,9
Mycotypha				•	,-
Rhizopus	55,2	61,9	48,5	45,1	30,9
ASCOMYCOTINA	40.0	00.7			
Chaetomium Sordaria	48,3	90,5	58,3	37,8	47,3
BASIDIOMYCOTINA	0	0	0,4	1,2	0
Ustilago	0	19,0	19.0	40.0	
DEUTEROMYCOTINA	v	19,0	12,8	48,3	44,4
Hyphomycetes, light					
Acremoniella	20,7	23,8	66,4	80,0	7.E A
Acrotheca	0	0	0	2,7	75,4
Acrostalagmus	0	Õ	0,9	0,5	5,8 1,0
Arthrobotrys	0	0	4,3	4,6	9,7
Cephalosporium	51,7	28,5	77,4	87,8	93,7
Fusarium	75,9	66,7	97,9	97,3	100,0
Fusidium	17,2	4,8	48,5	48,5	62,8
Gliocladium	13,8	9,5	29,8	48,3	26,6
Gliomastix	0	0	0	1,2	1,4
Gonatobotrys	0	0	3,0	1,0	4,8
Ostracoderma	17,2	4,8	26,4	12,7	20,8
Papulaspora	0	0	1,3	3,4	O.
Penicillium	96,9	100,0	78,3	82,7	74,9
Rhizoctonia	0	0	0	0,2	1,4
Sporotrichum Torula	24,1	14,3	8,5	20,7	9,7
Trichoderma	13,8	14,3	3,0	8,1	1,9
Trichothecium	6,9	4,8	8,9	14,6	9,2
Verticillium	$10,3 \\ 3,4$	14,3 0	33,6	19,5	25,6
Hyphomycetes, dark	υ, τ	U	0,9	0,7	1,4
Alternaria	100	100	100	99,8	100
Arthrinium	24,1	9,5	21,3	99,6 10,5	100
Aspergillus	$\frac{-2,1}{24,1}$	19,0	9,8	9,3	13,5 14,0
Botryotrichum	17,2	23,8	28,5	25,9	18,8
Botrytis	0	Ô	5,5	2,4	5,3
Cercospora			- /-	,-	0,0
Cladosporium	100	85,7	79,6	82,2	99,0
Curvularia	0	0	0,9	, O	0
Dactylella	0	0	0,4	0	0
Doratomyces	3,4	0	0	3,6	3,9
Drechslera	17,2	33,3	40,4	77,3	52,2
Epicoccum	58,6	9,5	30,6	35,4	44,0
Monodictys	0	9,5	1,3	1,2	1,4
Sepedonium	0	0	0	1,2	0,5
Stachybotrys	6,9	.0	1,7	1,7	1,4
Stemphylium	24,1	9,5	11,0	11,5	14,5
Trichocladium	37,9	28,5	13,6	17,1	19,3
Coelomycetes, Melanchoniales					
Colletotrichum	27,6	0	69,4	83,9	58,5
Coelomycetes, Sphaeropsidales	^	4.5			_
Phoma Septoria	0	4,8	0	2,4	2,4
Septoria OTHER FUNGI	. 0	0	2,1	6,6	1,0
BACTERIA, Streptomyces	6,9	0	31,9	29,8	38,6
PILOIDIUM, BUEDIUMIYCES	86,2	76,2	78,7	80,7	93,7

rather similar: Alternaria was the most frequent fungus  $(7.1-37\,^{0}/_{0})$ ; the second most frequent fungus was Cladosporium  $(5.3-42\,^{0}/_{0})$ ; and species of Fusarium were found on  $3.3-15.7\,^{0}/_{0}$  of the seeds examined. Of all oat seeds,  $42\,^{0}/_{0}$  were contaminated by

Cladosporium, 37 % by Alternaria and 15,7 % by Fusarium species (Table 6).

2. Fusarium species on various grains.

The prevalence of the Fusarium species in the seed samples varied greatly (Tables 7 and

Table 6. The frequence of various fungus genera and bacteria in cereal seeds in 1966-1971.

·	Rye	Winter	Spring wheat	Barley	Oats
	%	%	%	%	%
Number of seeds	457/5	9175	19000	7175	875
ZYGOMYCOTINA	10110	52.0	20000		
Mucor	2,2	0,7	8,0	0,6	0,7
	0,2	0,2	0,4	0,1	0,8
Mycotypha	2,9	2,0	0,8	0,8	0,8
Rhizopus	2,9	2,0	0,0	0,0	0,0
ASCOMYCOTINA	0	0,1	0,2	0,2	0,2
Chaetomium	+	0,1	+	+	0,2
Sordaria	T	U	7	Т-	U
DEUTEROMYCOTINA					
$Hyphomycetes,\ light$					
Acremoniella	0,3	0,2	0,4	0,7	3,0
Arthrobotrys	+	0,2	0,4	0,1	. 0
Cephalosporium	1,2	0,6	0,6	2,0	8,0
Fusarium	7,4	3,3	5,9	8,2	15,7
Gliocladium	0,3	0,1	+	0,4	0
Gonatobotrys	0,4	0,1	0,2	+	0
Oedocephalum	0	0	+	+	0
Papulaspora	0,1	0,1	+	0.1	1,0
Penicillium	6,5	9,9	12,6	3.2	4,5
Rhizoctonia	+	+	+	0,1	0,3
Torula	0,1	÷	÷	. +	. ,0
Trichoderma	0,1	0,2	÷	0.1	0,3
Trichoderma Trichothecium	0,4	0,2	1,2	0,1	1,0
	0,2	+	+	+	0,2
Verticillium	0,4	'	'	į	0,2
Hyphomycetes, dark	. 0.5	0.6	15,3	7,1	37,0
Alternaria	8,5	8,6		0,2	0 0
Arthrinium	0,3	0,6	2,0		
Aspergillus	0,1	0,2	0,2	0,2	0,3
Botryotrichum	0,7	- 0,1	0,5	0,8	5,0
Botrytis	0,7	0,7	0,4	0,2	0,3
Cercospora	. 0	0	0	0	0
Chlamydomyces	+	. +	0	0	0
Cladosporium	9,2	5,9	4,5	5,3	42,0
Doratomyces	-0	+	0	+	0
Drechslera	+	+	+	0	0,6
Epicoccum	2,1	1,9	5,9	2,0	6,7
Monodictys	0,7	0,5	+	0,7	0,2
Stachybotrys	+	<del>-</del>	+	0	0
Stemphylium	1,3	1,0	1,2	0,7	1,7
Trichocladium	0,2	0,1	0,2	0,2	0
	0,4	0,1	∨,=	٠,٠	·
Coelomycetes, Melanchoniales	0,8	+	0,6	+	0,5
Colletotrichum	υ,ο	I.	0,0	ı	0,0
Coelomycetes, Sphaeropsidales		^	0.1	11	, ,
Septoria	0	. 0	0,1	1,1	0
OTHER FUNGI	1,7	3,0	7,8	2,2	7,4
BACTERIA (Streptomyces)	2,0	2,4	2,2	3,0	2,6

Table 7. The frequence of Fusarium species in cereal samples in 1966—1971.

	Rye	Winter wheat	Spring	Barley	Oats
	%	wneat %	wheat %	Barrey %	%
	<u>.</u>				
Number of samples	183	367	759	287	35
SPOROTRIOHIELLA					
Fusarium chlamydosporum	0	1,6	2,1	0,7	0
» poae (& F. tricinctum)	19,7	24,8	29,8	59,2	100,0
» sporotrichioides	1,6	0,8	1,2	4,9	2,9
ROSEUM		•	•	•	,
Fusarium arthrosporioides	30,6	12,0	17,8	34,5	94,3
» avenaceum	19,7	17,2	20,4	41,8	100,0
ARTHROSPORIELLA	•	•	•	•	•
Fusarium semitectum	7,1	6,8	4,0	8,4	0
GIBBOSUM	•	•	•	•	
Fusarium acuminatum	1,1	9,5	9,0	6,3	0
» equiseti	1,1		1,6	0	
DISCOLOR			•		
Fusarium culmorum	33,9	28,1	54,8	86,8	100,0
» graminearum	3,3	6,0	5,0	12,5	100,0
» sambucinum	20,8	4,9	7,4	3,5	25,7
$^{ m w}$ tumidum	O	0	Ô	1,4	•
LATERITIUM				•	
Fusarium lateritium	0	0	0	1,7	0
LISEOLA				•	
Fusarium moniliforme	1,6	0	0	3,5	0
ELEGANS	•			•	
Fusarium oxysporum	23,5	9,3	9,5	15,7	11,4
» v. redolens	0	0	0,4	0,7	Ô
MARTIELLA			, i	,	
Fusarium coeruleum	1,6	0	0	0,7	0
» solani	3,3	3,3	3,3	16,0	5,7
Fusarium sp.	27,9	4,9	7,4	58,2	82,9

8). Certain species, however, seemed to be dominant in all the samples, regardless of their origin, year of harvest or type of cereal. In 1966—1971 the most frequent species was Fusarium culmorum (W. G. Smith) Sacc. which was found in on average of 60,7 % of all the samples and 1,1—12,1% of the seeds. The second most frequent species was F. poae (Pk.) Wr.  $(46.7)^{0}$  of samples and 0.8-5.9 0/0F. avenaceum (Fr.) Sacc., F. of seeds). arthrosporioides Sherb, and F. graminearum Schwabe were the next most frequent, also with very high contamination. The contamination of oats was especially high both in samples and on seeds.

In 1972 the exceptionally high temperatures and quite abundant local rainfall during the growing season were probably the main reason for the heavy fusarial infection in seeds (Uoti and Ylimäki 1974). The spring wheat in particular appeared to be quite heavily contaminated by Fusarium fungi (Table 9). The occurrence of various Fusarium species was again the same as in the material from 1966—1971, but the level of Fusarium culmorum contamination in spring wheat (30,9 %), barley (9,5 %) and oats (7,0 %) was exceptionally high, although the contamination by F. avenaceum, F. poae, F. tricinctum (Cda.) Sacc. and F. oxysporum (Schl. emend. Snyd. & Hans.) were also very high in all these cereals.

### 3. The microflora of various feedstuffs.

The microflora of 467 samples of various feedstuffs were investigated in 1967—1971.

Table 8. The frequence of Fusarium species in cereal seeds in 1966—1971

	Rye	Winter wheat %	Spring wheat %	Barley	Oats
		70			
Total number of seeds	4575	9175	18975	7175	875
SPOROTRICHIELLA			•		
Fusarium chlamydosporum	0	+	0,1	+	0
» poae (& F. tricinctum)	0,8	1,0	1,2	2,4	5,9
» sporotrichioides	÷	+	+	0,2	. 0,1
ROSEUM					•
Fusarium arthrosporioides	1,2	0,5	0,7	1,4	3,8
» avenaceum	0,8	0,7	0,8	1,7	4,0
ARTHROSPORIELLA	•	-			
Fusarium semitectum	0,3	0,3	0,2	0,3	0
GIBBOSUM	•				
Fusarium acuminatum	+	0,4	0,4	0,3	0
» equiseti	· +	0	0,1	0	0
DISCOLOR					
Fusarium culmorum	1,4	1,1	2,2	3,5	12,1
» graminearum	0,1	0,2	0,2	0,5	6,1
» sambucinum	0,9	0,2	0,3	0,1	1,0
» tumidum	0	0	0	+	0
LATERITIUM					_
Fusarium lateritium		. 0	0	0,1	0
LISEOLA		,			_
Fusarium moniliforme	+	0	0	0,1	0
ELEGANS					
Fusarium oxysporum	0,9	0,4	0,4	0,6	0,5
» » v. redolens	0	0	+	+	0
MARTIELLA					
Fusarium coeruleum	+	0	0	+	0
» solani	0,1	0,1	0,1	0,6	0,2
Fusarium spp.	1,1	0,2	0,3	2,3	3,3

Table 9. The percentage occurrence of Fusarium species in cereal seeds in 1972.

Fusarium s	pecies	Rye	Spring wheat	Barley	Oats
	Number of seeds	1200	13000	18000	10950
Fusarium	chlamydosporum	0	0	+	C
»	poae	2,3	3,4	3,9	8,2
»	tricinctum	0,2	1,5	6,2	2,4
»	sporotrichioides	0	+	+	C
»	arthrosporioides	0,2	1,5	1,2	1,0
»	avenaceum	2,1	7,4	7,3	5,9
»	semitectum	0,7	0,6	1,0	1,4
»	acuminatum	0	. 0,7	0,4	0,3
»	culmorum	1,3	30,9	9,5	7,0
»	graminearum	Õ	0,9	0,2	1,0
<b>»</b>	sambucinum	0	0,2	0,3	0,4
»	lateritium	0	÷	1,1	0,1
»	moniliforme	0	+	<u>.</u>	+
»	oxysporum	0,8	5,1	1,4	1,8
»	coeruleum	0	0	÷	Ć
»	solani	0	0,6	0,4	0,4
»	spp.	Ô	1,5	1,8	1,0

Table 10 shows the frequency of the different fungi found after incubation on wet filter paper. Penicillium, Mucor and Rhizopus were by far the most frequent fungus genera, but the occurrence of bacteria was also very significant.

4. List of fungus genera and species identified from the samples of cereal seeds and feedstuffs.

Myxomycetes, Physarales
Physarum Pers. em. Rostaf. sp.
Didymium Schrad. sp.

Zygomycotina, Mucorales
Helicocephalum Thaxt. sp.
Mucor Mich. ex Fr. spp.
Mycotypha microspora Fenner
Rhizopus nigricans Ehrenb.
Thamnidium elegans Link. ex S. F. Gray
Ascomycotina, Pyrenomycetes
Ceratocystis Ell. & Halst. sp.
Chaetomium elatum Kunze ex Fr.
Chaetomium globosum Kunze ex Fr.
Chaetomium olivaceum Cooke & Ellis
Chaetomium indicum Corda
Chaetomium fimicola Cooke
Chaetomium cochliodes Palliser

Table 10. The percentage occurrence of different microorganisms in feed stuffs.

Microorganism  ZYGOMYCOTINA	Feed grains	Commercial feed mixes	Нау	Silage	Total
				~	Total
Mucor	11,9	20,2	4,3	14.7	15,1
Rhizopus	14,0	14,5	5,1	10,4	12,9
ASCOMYCOTINA	,-	,•	٠,-	20,2	12,0
Chaetomium	2,3	5,3	7,0	3,6	4,4
DEUTEROMYCOTINA	,-	0,5	1,0	0,0	2,1
Hyphomycetes, light					
Acremoniella	1,9	0,1	51,0	. 0	1,4
Arthrobotrys	+	0,2	0,3	Ť	0,1
Cephalosporium	3,0	3,5	3,7	0,9	3,2
Fusarium	7,3	4,6	4,3	5,2	5,5
Gliocladium	0,5	0,2	+	0,5	0,3
Paecilomyces	0,4	1,7	0,8	4,0	1,2
Papulaspora	0,3	+	0,2	-, <u>,,</u>	0,2
Penicillium	15,2	13,8	10,6	13,4	13,9
Trichoderma	0,7	+	2,0	1,2	0,6
Trichothecium	0,8	+	4,9	0,1	0,9
Hyphomycetes, dark	•		-7-	*,-	-,-
Alternaria ·	9,3	2,1	10,0	1,1 '	5,5
Arthrinium	0,7	-,-	1,7	+	0,5
Aspergillus	3,6	1,9	7,9	5,2	3,4
Botryotrichum	0,2	0,3	+	0,2	0,3
Botrytis	0,3	+	0,8	Õ	0,2
Cladosporium	4,5	1,5	10,9	2,5	3,8
Doratomyces	0,4	0,1	0,6	0,3	0,3
Drechslera	0,3	+	0,3	0,0	0,2
Epicoccum	0,5	<u>;</u>	8,7	0	0,2
Stachybotrys	0,1	+	0,5	0,5	0,2
Stemphylium	0,2	+	0,3	+	0,1
Trichocladium	0,2	0,4	0,3	+-	0,3
Coelomycetes, Melanchoniales	0,4	U, x	0,0	1,	0,0
Colletotrichum	0,9	0,1	+	+	0,4
OTHER FUNGI	2,4	3,5	8,1	18,5	4,4
BACTERIA	4,4	٥٫٥	0,1	10,0	1,1
Streptomyces	7,4	7,9	7,3	4,2	7,5
Other	10,6	17,7	2,5	13,6	13,1

Chaetomium spinosum Chivers Melanospora zamiae Corda Melanospora fallax Zukal Sordaria fimicola (Rob.) Ces. & De Not Sordaria tetraspora Winter

Basidiomycotina

Septonema Corda sp.
Ustilago nuda (Jens.) Rostr.
Ustilago avenae (Pers.) Rostr.
Ustilago hordei (Pers.) Lagerh.

Deuteromycotina, Hyphomycetes, light Acremoniella atra (Corda) Sacc.

Acremoniella verrucosa Fogn.
Arthrobotrys superba Corda
Cephalosporium Corda sp.
Fusarium acuminatum (Ell. & Ev.) Wr.

Fusarium arthrosporioides Sherb.

Fusarium avenaceum (Fr.) Sacc.

Fusarium chlamydosporum Wr. & Rg.

Fusarium coeruleum (Lib.) Sacc.

Fusarium culmorum (W. G. Sm.) Sacc.

Fusarium dimerum Penz.

Fusarium equiseti (Cda.) Sacc.

Fusarium graminearum Schwabe

Fusarium lateritium Nees

Fusarium moniliforme Sheld.

Fusarium nivale (Fr.) Ces.

Fusarium oxysporum Schl. emend. Sny-

der & Hansen

Fusarium oxysporum Schl. emend. Snyder & Hansen var redolens (Wr.) Gordon Fusarium poae (Pk.) Wr.

Fusarium semitectum Berk. & Rav.

Fusarium sambucinum Fuckel

Fusarium sambucinum Fuckel forma 6 Wr. Fusarium sambucinum Fuckel var. coeruleum Wr.

Fusarium solani (Mart.) App. & Wr. emend.

Snyder & Hansen

Fusarium sporotrichioides Sherb.

Fusarium tricinctum (Cda.) Sacc.

Fusarium tumidum Sherb.

Fusarium Link. ex Fr. spp.

Fusidium Link. ex Fr. sp.

Gliocladium roseum (Link.) Bain.

Gliomastix murorum (Corda) Hughes

Gonatobotrys Corda sp.

Oedocephalum Preuss sp.

Ostracoderma Fr. sp.

Papulaspora Preuss sp.

Penicillium Link. ex Fr. spp.

Pullularia pullulans (de Bary) Berkh.

Rhizoctonia solani Kühn

Sporotrichum Link. ex Fr. sp.

Torula herbarum (Pers.) Link. ex S. F. Gray

Trichoderma viride Pers. ex Fr.

Trichothecium roseum Link. ex Fr.

Verticillium Nees ex Wallr.

Deuteromycotina, Hyphomycetes, dark

Alternaria tenuis Nees

Alternaria cheiranthi (Fr.) Bolle

Alternaria tenuissima (Kunze ex Pers.) Wiltsh.

Alternaria raphani Groves & Skolko

Arthrinium sphaerospermum (Corda) M. B. Ellis

Aspergillus flavus group

Aspergillus fumigatus group

Aspergillus glaucus group

Aspergillus niger group

Aspergillus Mich. ex Fr. sp.

Bispora Corda sp.

Botryotrichum Sacc. & March sp.

Botrytis cinerea Pers. ex Fr.

Cercospora Fres. sp.

Chlamydomyces palmarum (Cooke) Mason

Cloridium Link sp.

Cladosporium herbarum (Pers.) Link ex Fr.

Cladosporium cladosporioides (Fres.) de Vries

Curvularia Boedijn sp.

Dactylella Grove sp.

Doratomyces microsporus (Sacc.) Morton et

G. Smith

Doratomyces purpureofuscus (Fr.) Morton

et G. Smith

Doratomyces nanus (Ehrenb. ex link.) Mor-

ton et G. Smith

Doratomyces stemonitis (Pers.) Link. ex Fries

Drechslera avenae (Eidam) Scharif Drechslera graminea (Rabenh. ex Schlecht.) Shoem

Drechslera teres (Sacc.) Shoem

Epicoccum purpurascens Ehrenb. ex Schlecht.

Schlecht.

Geotrichum candidum Link.

Gilmaniella Barron sp.

Monodictys levis (Wiltsh.) Hughes

Sepedonium Link ex Fr. sp.

Stachybotrys atra Corda

Stachybotrys aurantia Barron

Stemphylium botryosum Simmons

Stemphylium consortiale (Thüm.) Groves & Skolko

Trichocladium asperum Harz.

Deuteromycotina, Coelomycetes, Melanchoniales

Colletotrichum graminicola (Ces.) Wils. Deuteromycotina, Coelomycetes, Sphaeropsidales

Ascochyta graminicola Sacc. Ascochyta hordei Hara Asteroma DC. ex Fr. sp. Phoma sensu Sacc. sp. Pyrenochaeta de Not. sp. Septoria nodorum Berkeley Septoria tritici Roberge

### DISCUSSION

Most studies on the microflora of cereal seeds deal with stored grains, with pathogenic fungus species or with certain fungus genera or species only. More comprehensive surveys on the microflora of ripening cereal crops standing in the field or at harvest time are rather few (Spicher 1958, Malone and Muskett 1964, Koroleva 1967, Noble and Richardson 1968, Ylimäki 1970, Sodnomdorzh 1973, Flannigan 1970, 1974). We know, however, that the cereal crops in the field are regularly contaminated with a variety micro-organisms i.e. fungal species (the »field fungi»), bacteria, and also insects.

When the condition of seeds is examined after incubation, the picture is often that of a rich fungus flora mostly consisting of saprophytes or weak parasites which apparently have little influence on seed quality. However, these fungi cannot be ignored because the specific composition of this flora may give some indications of the condition of the seeds. Furthermore, a knowledge of the general fungus flora is also of importance when samples for fodder and industrial purposes are examined, because mould on

grains and feedstuffs carries with it the risk of mycotoxin contamination (Ciegler, Kadis and Ajl 1971, Kadis, Ciegler and Ajl 1971, 1972, Palti 1978, Ylimäki et. al. 1979). For this reason the occurrence of both pathogenic and saprophytic fungi of cereal seeds and feedstuffs was recorded during this survey.

The quantity of fungi varied annually very much according to the climatic conditions during the growing season, especially during the harvest time. On the other hand, the selection of fungi was principally the same in all years.

In cereals, Alternaria species were by far the most frequent fungi, and occurred in more than 90 % of all samples: the contamination of seeds varied between 7.1 and 37 %. The next most frequent fungus genera were Cladosporium, Fusarium and Penicillium, occurring in 73—100 %, 56—100 % and 47—100 % of samples, respectively, and on seeds at rates of 5,3—42 %, 3,3—15,7 % and 3,2—12,6 %, respectively.

Infection by Fusarium species was particularly dependent on the moisture conditions of the growing season (cf. Hewett 1950, Gor-

don 1952, Konszky and Pasty 1971). The relative high humidity in, e.g., 1967 and 1972 was the main reason for the heavy fusarial infection in cereal seeds in both these years (Uoti and Ylimäki 1974).

The rather abundant occurrence of Fusarium fungi is worthy of mention because some species are known to produce mycotoxins in Finland (Ylimäki et. al. 1979). The most common Fusarium species were F. culmorum, F. avenaceum, F. arthrosporioides and F. graminearum but contamination by F. poae, F. tricinctum and F. oxysporum was also very high in all cereals.

A fact to be taken in to special consideration was the very scanty occurrence of Fusarium nivale on seeds. It was found only in very few cases on the seeds of rye in all years studied, despite the special attention

paid to its occurrence. On the other hand, snow mould damage caused by F. nivale is a very serious problem in Finland because it restricts or hinders the cultivation of winter cereals. Jamalainen (1962) has reported that seed treatment often but not always provides good control of snow mould. The treatment of stands with certain fungicides in autumm is a very good method for preventing snow mould (Jamalainen 1964). This being the case, it seems that the seed-borne infection of cereals by F. nivale has scarcely any decisive importance as a cause of snow mould compared with soil infection (as reported also by Sprague 1950, Colhoun 1970 and Taylor 1970).

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### SELOSTUS

### Viljan jyvien ja joidenkin rehujen sienistö

### AARRE YLIMÄKI

### Maatalouden tutkimuskeskus

Vuosina 1966—1973 tutkittiin sienistö eri tahoilta maata peräisin olleista 2601 rukiin, syysvehnän, kevätvehnän, ohran ja kauran jyvänäytteestä. Yleisimmät jyvistä tavatut sienisuvut olivat Alternaria, Cladosporium, Fusarium ja Penicillium, joita esiintyy 90—100, 73—100, 56—100 ja 47—100 prosentissa näytteistä. Tavallisimmat Fusariumlajit olivat F. culmorum, F. poae, F. tricinctum, F. avenaceum, F. arthrosporioides, F. graminearum ja F. oxysporum. Viljojen oraslumihomeen aiheuttajana tunnettua F. nivale-lajia tavattiin vain muutamista jyvänäyteistä.

Rehuvilja-, rehuseos-, heinä- ja säilörehunäytteissä, joita tutkittiin yhteensä 467 kpl, olivat tavallisimmin tavatut sienet Penicillium, Mucor ja Rhizopus, joskin verraten yleisiä olivat myös Alternaria, Fusarium, Chaetomium, Aspergillus ja Cladosporium -sienet. Bakteereista varsinkin sädesienet olivat yleisiä.

Tuoreena säilötyssä viljassa, josta ilma oli onnistuttu poistamaan, sieniä ei joko tavattu lainkaan tai niitä oli erittäin vähän.

Kaikkiaan viljoista ja rehuista määritettiin 113 eri sienilajia tai -sukua.

### Seria PHYTOPATHOLOGIA N. 77 — Sarja KASVITAUDIT n:o 77

### EXPERIMENTS WITH NON-MERCURY SEED DRESSINGS ON SPRING CEREALS

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Vanhanen, R. 1981. Experiments with non-mercury seed dressings on spring cereals. Ann Agric. Fenn. 20:89—101. (Agric. Res. Centre, Inst. Pl. Path. SF-01300 Vantaa 30, Finland.)

Since the mid-1960s, official efficiency tests have been performed for the investigation of dozens of non-mercury seed dressings in the control of seed-borne diseases of spring cereals. Their efficacy against leaf stripe of barley, loose smut of barley and of wheat, stinking smut of wheat, and loose smut of oats has been investigated in field and greenhouse experiments. Most of the preparations have been highly effective against stinking smut and against loose smut of oats, while few of them have produced good results in the control of stripe disease or of loose smut of barley and of wheat.

Most of the investigated seed dressings have had to be rejected on account of narrow range of effect, while others have been screened out for toxicological reasons or phytotoxicity. Through the combination of active ingredients, it has proved possible to improve the effects of the preparations on various species of fungi, though no seed dressing agent has been adopted as yet that is highly effective against the smut diseases as well as the stripe disease. Four non-mercury seed dressings have been sales licensed in Finland for use on spring cereals: Benlate against loose smut of oats and stinking smut of wheat, Panoctine Plus against stripe disease and stinking smut, and Vitavax and Vitavax T-liquid against loose smut of barley and of wheat, stinking smut of wheat and loose smut of oats.

Index words: Testing of fungicides, seed dressings, barley, oats, spring wheat, Drechslera graminea, Ustilago nuda, U. avenae, U. tritici, Tilletia caries.

### INTRODUCTION

Organomercury compounds have been used in Finland for the control of plant diseases in cereals since the late 1920s. They have been superior to other agents. The mercury compounds are effective against most seed-borne pathogens and are cheap. They are not

phytotoxic and they are available both in powder and in liquid forms, highly suitable for use in various kinds of seed treater.

Countering their good features, the mercury compounds possess drawbacks too. They are ineffective against loose smut of barley

and of wheat and do not protect the sprouts against contamination from the soil. But the most serious problem is their toxicity, which is a hazard both to persons performing the dressing and to wildlife. Since the risks ensuing from mercury were recognized in the 1960s, there have been efforts to reduce the utilisation of mercury seed dressings and replace them with less toxic preparations. Today many countries have totally prohibited or severely restricted the use of mercury seed dressings. In Finland the use of poorly degradable methyl mercury compounds was abandoned in 1969, and the use of the more pro-environmental ethyl mercury preparations exclusively was adopted.

The finding of effective non-mercury seed

dressings has proved to be more difficult than was expected. Only the development of systematically-effective active ingredients and the combination of various ingredients into a mixture has brought within sight preparations that can compete on equal terms with the mercury compounds. The effect of nearly 100 non-mercury preparations against plant diseases occurring in cereals has been investigated at the Institute of Plant Pathology in the past 15 years. This report brings together the results mainly of preparations that are already in use for the control of plant diseases in some countries, or that seem to be promising alternatives to mercury seed dressings.

### MATERIAL AND METHODS

In control tests on leaf stripe of barley and on loose smut of barley and of wheat, use was made of batches of seed which were infected naturally and as heavily as possible. In the tests on stinking smut of wheat and loose smut of oats, healthy and well-germinating seeds were artificially inoculated with smut spores.

In the inoculation with loose smut of oats, 250 g of oat seed and 1—3 g of smut spores are mixed into a nutrient solution containing, per litre of water, two grammes each of  $(NH_4)_2SO_4$ ,  $K_2SO_4$  and  $KH_2PO_4$  as well as one g each of glucose,  $MgSO_4$ , NaCl and CaCl<sub>2</sub>. By vacuum suction, air is drawn from the mixture in a dessicator for 20 minutes, whereafter the air is slowly allowed to be reabsorbed, whereon the spores are carried in between the grain and the coat. The grains are then dried on blotting paper at room temperature. After two days they are placed in a glass jar lined with damp blotting paper at a temperature of  $+25^{\circ}C$  for 24 hours, for

the spores to germinate. After drying, the seeds are ready to be dressed.

Wheat grains are inoculated with dry spores of stinking smut by shaking a mixture of seeds and spore dust in a glass jar for 5—10 minutes. From 5—8 g of dust is used per kg of seed. After inoculation, the grains can immediately be dressed.

When the dressing was done with powdery preparations, the desired quantities of seed and dressing were weighed into a cylindrical glass jar. Until 1975 the liquid preparations were pipetted on to the inner surface of a glass vessel containing seeds. In both cases, the mixing has been performed by violent manual shaking or mechanical rotating of the vessel for five minutes. Since 1976 a device has been used in liquid dressing in which the dressing liquid is sprayed by compressed air, in the form of a fine mist, in among grain being mixed in a rotating glass jar (Vanhanen 1977).

Control tests with leaf stripe of barley

# SEED DRESSINGS USED IN THE TESTS

Mercury compound		
Ceresan	powder	2-methoxyethylmercury silicate, Hg 1,5%
Non-mercury compounds		
Baitan F-powder and B.Fliquid Bas 3302 F BAS 389 01 F BAS 395 03 F	powder and liquid powder liquid liquid	triadimenol 15 %, fuberidazole 2 % 25-dimethyl-N-cyclohexyl-3-furancarbonic amide 50 % maneb 32 % 2,5-dimethyl-N-cyclohexyl-N-methoxy-3-furancarbonic amide 50 % 2,5-dimethyl-N-cyclohexyl-N-methoxy-3-furancarbonic amide 45 %,
Bayer 5488 Bayer 5590 Bayer 6743 and Bayer 6744 Benlate	powder liquid powder and liquid powder	imazalil 2,5 % ohexachlorbenzene 20 % fuberidazole 3 % ohexachlorbenzene 20 % fuberidazole 3 % ohexachlorbenzene 20 % ohexachlorbenzene 25 % ohexachlorbenzene 4 % ohexachlorbenzene 56 % ohexachlorbenzene 5
Busan 72, KVK/Busan, TCMTB'30 EC Derosal Dithane M-45 Dithane Z-75 Dichane Z-75	liquids powder powder	2-(thiocyanomethylthio)benzothiazole 60 %, 50 %, 40 %, respectively carbendazim 60 % mancozeb 80 % zinc-ethylenebisdithiocarbamate 65 %
Folcidin Fungaflor-powder and Fliquid Granosan	ngun powder powder and liquid powder	carbendazim 6 % cypendazole 50 % imazalii 3,3 % carbendazim 15 % maneh 60 %
Panoctine Panoctine Plus Panoctine Universal PL 3338	liquid liquid liquid liquid	quazatine 35 % imazalil 2 % fenturam 8 % thisbendazole 1.5 % thintoxene 20 %
PL 3417 PL 3418 Pomarsol Forte, Scorvine Rovral	liquid liquid <b>powders</b> powder	thiabendazole 2%, quintozene 3%, carboxin 5% thiabendazole 2%, quintozene 5%, carboxin 5% thiram 80% and 50%, respectively inrodione 50%.
Sidipreg Sidipreg 77 Sisthane Topsin M Trimangol	liquid liquid liquid powder	thiabendazole 2 %, carboxin 3 %, sorbatoxin 10 % thiabendazole 2 %, carboxin 5 % phenapronil 24 % thiophanate-methyl 70 % maneb 80 %
	inquid powder liquid powder liquid powder powder and liquid	nuarumol 10 % carboxin 75 % thiram 15,4 % carboxin 175 % thiram 15,4 % carboxin 17,3 %, thiram 15,4 % fuberidazole 3 %, hexachlorbenzene 20 % fuberidazole 0,5 %, sodium dimethyldithiocarbamate 30 % fuberidazole 3 %, quintozene 25 % imazalil 2 % carbendazim 20 % carboxin 10 % carboxin 10 % carboxin 10 %
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have been performed both in greenhouse and in the field. In greenhouse tests the seeds have been sown into sterilised soil in sprouting pots and allowed to sprout at a temperature of +10°C, whereafter the plants have been transferred into the greenhouse. The analysis has been done 5-6 weeks after sowing, by counting the intact specimens and those contaminated with leaf stripe. The field trials have been done with a Planet Jr. seeder or with an Øyjord plot drill, usually in plots of 10 square metres with four replicates. The healthy and the leaf striped plants were counted at the time when the barley ears emerged, at a distance of  $4 \times 2$  metres of row per plot.

The control tests with loose smut of barley and of wheat have all been field trials and have been established in the same way as the tests on leaf stripe of barley. The number of ears was counted at a distance of  $4\times0.5$  metres of row per plot and the smutted ears were counted throughout the entire plot.

The tests with loose smut of oats and stinking smut of wheat have been done in boxes 6 dm<sup>2</sup> (of 60 grains each with five replicates). The former were allowed to sprout at room temperature, and the latter at a temperature of  $+10^{\circ}$ C. The sprouts have been transplanted when they were five centimetres high, to grow in furrows made in the field. The cereal, pulled out of the soil in autumn, has been analysed by counting the healthy and the smutted specimens.

### RESULTS AND DISCUSSION

# Efficacy of seed dressings against plant diseases

Leaf stripe of barley (Drechslera graminea, Fig. 1). The results of the control tests on leaf stripe of barley are shown in Tables 1 and 2. Most effective among the non-mercury seed dressings against leaf stripe was Sisthane, which contains phenapronil as active ingredient. It was even more efficient than mercury. But tests with Sisthane had to be discontinued for toxicological reasons. Another effective compound against leaf stripe is imazalil, which is the active ingredient in preparations including Fungaflor, Panoctine Plus, Panoctine Universal, 7118/1A and 9051. Of the above-mentioned preparations Panoctine Plus became in 1980 the first non-mercury seed dressing to be allowed a sales license in Finland for the control of leaf stripe. With normal amounts of use, the effect of the other agents fell short of 90 per cent.

Insufficient effect against leaf stripe happens to be a threshold on which non-mercury preparations often fall. This is the case particularly in Finland, where agents such as maneb, mancozeb, thiocyanomethylthiobenzothiazole and carboxin, with which good results in the control of leaf stripe disease have been achieved in many countries, have proved to be inadequately effective. So far, only imazalil seems to provide an acceptable alternative to mercury compounds in the control of leaf stripe. Both powder and liquid applications can be prepared of this. A disadvantage is the non-existent effect against the smut diseases, and it is consequently used in mixtures together with other active ingredients.

Loose smut of barley and of wheat (Ustilago nuda and U. tritici, Figs. 2 and 3). Mercury preparations are completely ineffective against loose smut of barley and loose smut of wheat. Vitavax, with carboxin as its active ingredient, was used as reference prod-

Table 1. Effect of seed treatments on leaf stripe of barley, field trials, 1972-80.

	No. of	Rate per	Proporti emergence	onals for (Untr. = 100)	Untreated	Per cen	t effect
Treatment	expt.	100 kg seed	non- mercury	mer- cury	% attack	Per cent non- mercury 1  32,6 85,0 54,0 6,5 14,6 81,2 58,5 61,3 73,9 —11,8 94,4 97,4 26,9 10,8 86,8 98,4 47,2 99,6 100 24,3 —30,5 —13,1 59,5 32,6	mercury
Mercury compound							
Ceresan	.8	200 g		109	29,2		99,1
Non-mercury compounds							100
Baitan F-liquid	1	200 ml	99	104	9,2		100
BAS 3302 F	2	200 g	66	79	30,1	•	99,9
	1	300 g	90	111	25,0		98,8
Bayer 6743	2	200 g	99	113	47,2		98,7
Bayer 6744	<b>2</b>	200 ml	93	113	47,2		98,7
Benlate + Pomarsol F	1	50  g + 100  g	56	65	39,8		99,7
	1	100 g + 100 g	57	65	39,8	,	99,7
Benlate $+$ Trimangol	1	50  g + 100  g	57	65	39,8		99,7
2011210	1	100  g + 100  g	84	92	20,3		100
Derosal	4	200 g	89	100	39,8	11,8	99,0
Fungaflor-powder	2	300 g	101	107	16,2	94,4	98,8
Fungaflor-liquid	5	300 ml	91	110	30,4	97,4	98,8
Granosan	3	200 g	108	112	39,8	26,9	98,7
Panoctine	1	200 ml	92	94	58,4	10,8	98,8
Panoctine Plus	3	200 ml	96 -	109	38,9	86,8	99,0
Panoctine Universal	$\dot{\tilde{2}}$	200 ml	104	107	16,2	98,4	98,8
Rovral	1	200 g	92	102	22,3	86,5	99,6
Sidipreg	1	300 ml	113	111	25,0	28,4	98,8
Sidipreg	î	400 ml	85	65	39,8	47,2	99,7
Sisthane	$ar{2}$	200 ml	101	103	15,8	99,6	99,8
Sistilane	1	320 ml	90	111	10,1	100	98,0
TCMTB 30 EC	$\hat{3}$	200 ml	83	102	33,6	24,3	99,0
Topsin M	i	50 g	83	92	20,3	-30.5	100
Topsiii w	1	250 g	49	65	39,8	13,1	99,7
Trimidal 10 S	3	200 ml	92	106	13,9		99,2
Trimidal 10 S	1	250 ml	99	104	9,2		1Ó0
Vitavax T-liquid	. 4	300 ml	94	102	30,8	45,2	99,2
Vitavax 1-liquid Voronit-liquid	4	300 ml	99	100	30,3	33,9	99,3
voronit-liquid 7118/1A	3	200 g	94	106	13,9	99,1	99,2
	2	200 g	97	108	9,7	99,0	99,0
9051/1	1	200 g 200 ml	95	104	9,2	93,5	100
9051/3A	$\frac{1}{2}$	300 ml	94	107	16,2	92,9	98,8

uct in the tests on loose smut (Tables 3 and 4). Vitavax was granted a sales license in Finland, for the control of smut diseases, in 1971. Its effect against loose smut of barley is almost complete. Against loose smut of wheat, Vitavax has usually had a very good effect but the years 1978 and 1979 are exceptions, for then the effect did not exceed 80—85 per cent. Apart from the high price, the powdery consistency of Vitavax has restricted its use. For this reason Vitavax T-liquid (with carboxin and thiram as active ingredients), which is effective against loose smut

of barley, was put on the market in 1979. Vitavax T-liquid is no longer recommended for the control of loose smut of wheat, because in some tests the effect has not exceeded 60 per cent.

The most promising among other active ingredients proved to be triadimenol, which is the ingredient effective against smut fungi in Baitan F preparations. Against loose smut of barley, Baitan F powder and liquid dressing agents are slightly inferior to Vitavax but all the more effective in the control of loose smut of wheat.



Fig. 1. Leaf stripe of barley (Drechslera graminea).



Fig. 2. Loose smut of barley (Ustilago nuda).



Fig. 3. Loose smut of wheat (Ustilago tritici).



Fig. 4. Stinking smut of wheat (Tilletia caries).



Fig. 5. Loose smut of oats (Ustilago avenae).

Stinking smut of wheat (Tilletia caries, Fig. 4). Stinking smut is one of the most easily controlled diseases of cereals (Table 5). Almost all the non-mercury seed dressings tested had excellent effects against stinking smut, the effect of most of them being 100 per cent. In addition to the Vitavax preparations, sales licenses have also been given for Panoctine Plus, Benlate (1971) and Voronit

(1972). The sales license for the Voronit preparation was withdrawn in 1977, when hexachlorbenzene, which was contained in it, was found in experiments on animals to have caused cancer.

Loose smut of oats (Ustilago avenae, Fig. 5). Loose smut of oats cannot be completely controlled with mercury compounds, even if the quantity applied to oats is 50 per cent higher

Table 2. Effect of seed treatments on leaf stripe of barley, greenhouse trials, 1965-79.

	No. of	Rate per	Proportion emergence (	onals for Untr. = 100)	_	Per cen	teffect
Treatment	expt.	100 kg seed	non- mercury	mer- cury	Untreated % attack	non- mercury	mercury
Mercury compound				100			00.0
Ceresan	31	200 g		103	21,7		99,3
Non-mercury compounds			0.77	101	14 5	21.0	91.7
Baitan F-powder	1	200 g	97	101	14,5	$31,0 \\ 42,3$	100
Baitan F-liquid	1	200 ml	92	94	33,3	$\frac{42,3}{77,3}$	100
BAS 3302 F	1	200 g	104	107 104	30,9 49,8	100	100
	1	300 g	96	104	28,2	27,1	100
Bayer 5488	3	200 g	100		,	92,0	100
Bayer 5590	4	1000 ml	92	102	27,0		100
Bayer 6743	3	200 g	102	105 105	49,1	54,1 60,6	100
Bayer 6744	3	200 ml	95		49,1	20,7	99.5
Benlate	7	200 g	100	106	12,0	,	98,3
Benlate $+$ Dithane M-45	2	100 g + 100 g	105	106	10,7	79,6	100
Benlate + Trimangol	1	100 g + 100 g	103	107	30,9	88,0	98,8
	3	125 g + 125 g	108	104	17,7	91,9	99,1
Busan 72	4	65 ml	102	103	8,1	63,8	
	<b>2</b>	100 ml	104	108	14,8	87,6	98,5 100
KVK Busan	4	200 ml	99	103	5,7	83,8	100
Derosal	5	200 g	99	`105	45,6	1,9	100
Dithane M-45	• 5	200 g	101	101	26,1	69,3	100
Folcidin	1	150 g	96	104	49,8	17,9	95,9
Fungaflor-powder	2	300 g	106	105	23,4	98,2	98,6
Fungaflor-liquid	6	300 ml	102	105	40,6	99,6	100
Granosan	4	200 g	103	105	49,3	83,0	100
Panoctine	1	200 ml	103	104	52,6	24,0	100
Panoctine Plus	4	200 ml	105	106	44,9	95,8	95,9
Panoctine Universal	2	200 ml	105	105	23,4	88,8	100
Rovral	1 ,	200 g	. 101	109	32,2	71,4	100
Scorvine	1	200 g	104	103	31,1	51,4	100
Sidipreg	1	200 ml	106	102	31,8	49,1	100
	2	300 ml	101	106	40,4	65,6	100
	1	400 ml	133	120	17,1	85,4	100
Sisthane	2	200 ml	104	102	32,8	100	
	1	320 ml	97	101	14,5	100	91,7 100
TCMTB 30 EC	5	200 ml	101	105	41,4	88,4 19,2	100
Topsin M	2	50 g	100	111	24,5	2,6	100
	1	150 g	100	107	30,9	,	100
Trimangol	5	150 g	96	101	26,1	86,5 49,6	97,2
Trimidal 10 S	. 3	200 ml	101	101	26,7		100
Vitavax	6	200 g	102	103	5,8 14.5	75,0 61,1	98,5
_	2	250 g	104	102	14,5	89,0	100
Vitavax T-liquid	5	300 ml	103	106	41,5	36,5	100
Voronit	3	200 g	101	101	28,2		100
Voronit-liquid	5	300 ml	105	108	38,5	74,8	98,9
	4	500 ml	101	101	17,5	69,8 92,9	97,2
7118/1A	3	200 g	103	101	26,7		95,9
9051/1	2	200 g	103	98	23,9	91,7	
9051/3 <b>A</b>	1	200 ml	107	94	33,3	92,8	100
	2	300 ml	101	105	23,4	69,7	95,9

than it is for other cereals (Table 6). The effect of the mercury seed dressings has been 60—99 per cent, depending on the year. Apart from a few exceptions, non-mercury prepa-

rations have had a good effect against this loose smut. Most surprising is the non-existent effect of Panoctine Plus and Voronit in the control of loose smut of oats, although

Table 3. Effect of seed treatments on loose smut of barley, 1969-80.

Treatment	No. of	Rate per	Proporti heads (U	ionals for ntr. = 100)	Untreated — % smut-	Per cer	nt effect
	expt.	100 kg seed	prod- uct	refer- ence prod- uct	ted heads	prod- uct	refer- ence prod- uct
Vitavax (reference product)	9	200 g		98	7,6		98,4
Baitan F-powder	4	200 g	99	101	11,7	95,0	99,2
Baitan F-liquid	3	200 ml	108	105	14,6	95,2	98,6
BAS 3302 F	1	200 g	93	81	9,1	95,6	97,8
	1	300 g	95	106	0,8	98,9	100
Benlate + Pomarsol F	1	50  g + 100  g	90	81	9,1	49,5	97,8
	1	100  g + 100  g	95	81	9,1	30,8	97,8
Benlate + Trimangol	1	50  g + 100  g	90	81	9,1	18,7	97,8
	1	100  g + 100  g	97	103	4,2	81,0	92,9
Derosal	2	200 g	97	94	5,0	37,0	98,9
Fungaflor-liquid	1	300 ml	106	106	0,8	15,8	100
Granosan	1	200 g	105	106	0,8	1,7	100
Panoctine Universal	3	200 ml	95	96	8,3	71,3	99,3
Rovral	1	200 g	96	96	7,5	-4,0	99,1
Sidipreg	1	200 ml	98	81	9,1	38,5	97,8
	1	400 ml	91	103	4,2	50,0	92,9
Sisthane	1	200 ml	105	107	16,1	55,3	98,1
	2	320 ml	89	96	8,8	81,2	99,4
TCMTB 30 EC	2	200 ml	96	94	5,0	6,9	98,9
Topsin M	1	50 g	97	103	4,2	Ô	92,9
	1	250 g	100	81	9,1	11,0	97,8
Trimidal 10 S	4	200 ml	101	99	10,3	60,4	99,0
	1	250 ml	100	103	13,1	98,5	99,9
Vitavax	. 1	100 g	111	97	0,1	100	1Ó0
Vitavax T-liquid	4	300 ml	108	103	9,4	94,4	99,3
Voronit-liquid	1	300 ml	115	106	0,8	<b>— 2,9</b>	100
7118/1A	4	200 g	102	99	10,3	24,1	99,0
9051/1	4	200 g	103	101	11,7	78,7	99,2
9051/ <b>3A</b>	2	200 ml	107	105	14,6	80,2	99,0
	3	300 ml	97	96	. 8,3	92,9	99,3

their effect is extremely good against stinking smut of wheat. Sales licenses, for the control of loose smut of oats, have so far been issued for three non-mercury seed dressing agents: Vitavax, Vitavax T-liquid and Benlate.

# Effect of seed dressing agents on sprouting and yield

Tables 1—6 show the percentages of effect and also the effect of the preparation on the number of specimens. In the tests on stinking smut of wheat, loose smut of oats and in the greenhouse tests on leaf stripe of barley, use was made of the same kind of soil in

every treatment each year; all the developed specimens were counted and, too, the seed in the smut tests was of perfect quality, so it follows that the results of the effects of the preparations upon sprouting were more reliable than in the field trials. Most of the preparations in these tests caused a slight increase in sprouting, as compared with nontreatment. The improvement with non-mercury preparations, however, was lower on average than it was with the mercury preparations.

In the field trials the numbers of specimens were usually higher on the untreated plots than on the plots planted with dressed

Table 4, Effect of seed treatments on loose smut of wheat, 1968-80.

	No. of	Rate per	Proporti heads (Ur	onals for $tr. = 100$	Untreated — % smut-	Per cen	t effect
Treatment	expt.	100 kg seed	prod- uet	refer- ence prod- uct	ted heads	prod- uct	refer- ence prod- uct
Vitavax (reference	12	200 g		96	4,7	0.4.0	91,7
Baitan F-powder	5	.200 g	96	99	6,1	94,9	86,0
Baitan F-liquid	3	200 ml	96	103	4,7	95,9	87,1
BAS 3302 F	1	200 g	107	102	2,2	86,4	95,5
Bayer 6743	$ar{2}$	200 g	106	91	7,1	92,3	97,2
Bayer 6744	$\bar{2}$	200 ml	94	91	7,1	94,5	97,2
Benlate + Pomarsol F	1 .	50 g + 100 g	113	102	2,2	63,6	95,5
Beniate + Folharson F	î '	100 g + 100 g	100	102	2,2	95,5	95,5
Davidata   Trimongol	1	50 g + 100 g	102	102	2,2	27,3	95,5
Benlate + Trimangol	1	100 g + 100 g	106	96	2,2	81,8	95,5
- ·	3	. 200 g	94	94	5,5	72,7	96,6
Derosal	1	200 g	95	81	7,3	61,6	97,2
Granosan	$\frac{1}{2}$	200 g 200 ml	106	91	7,1	2,3	97,2
Panoctine	$\overset{2}{2}$	200 ml	103	91	7,1	4,5	97,2
Panoctine Plus	2 3	200 ml	102	96	7,6	60,6	89,2
Panoctine Universal		200 mi	99	102	6,5	<b>— 8,8</b>	98,9
Rovral	1		98	99	2,2	2,3	95,5
Sidipreg	2	200 ml	93	96	5,7	77,7	86,7
Sisthane	3	200 ml	94	93	8,2	89,3	84,4
	2	320 ml		101	4,6	-74.8	96,3
TCMTB 30 EC	2	200 ml	79	96	$\frac{4}{2},0$	9,1	95,5
Topsin M	1	50 g	100		$^{2,2}_{2,2}$	4,5	95,5
_	1	200 g	105	102		92,3	85,8
Trimidal 10 S	5	200 ml	87	95	6,7	100	100
	1	250 ml	13	122	3,6	98,2	92,9
Vitavax	2	100 g	105	86	0,2	,	92,9
Vitavax T-liquid	7	300 ml	102	99	5,3	78,8	
Voronit-liquid	1	300 ml	104	100	6,9	13,0	97,1
7118/1A	5	200 g	99	95	6,7	60,6	85,8
9051/1	5	200 g	98	99	6,1	45,4	86,0
9051/3A	3	200 ml	104	103	4,7	57,1	87,1
3001/011	3	300 ml	94	96	7,6	77,9	89,2

seed, although the differences were not notable. In assessing the phytotoxicity of the preparations, too much weight should not be given to the results of the field tests, as the count was done at the stage of ear emergence only for a small part of the plot, and it was found that the variation between different replicates and between different parts of plots was extremely high. It was also noticed in the tests on loose smut that tillering was greater in plants contaminated by smut, which were also more numerous precisely in the untreated plots.

To ascertain any phytotoxicity, greenhouse

tests were made on the best of the preparations. In these tests the seed was dressed also with greater than normal amounts of the agents. A clear phytotoxic effect was observed in some species or varieties of cereal treated with preparations containing thiocyanomethylthiobenzothiazole, nuarimol and phenapronil.

The yield results of the field trials on loose smut of barley and loose smut of wheat and on leaf stripe of barley are shown in Table 7. The yield loss caused by these diseases is usually estimated to be nearly as great as the percentage of diseased plants or ears. In

Table 5. Effect of seed treatments on bunt of wheat, 1965-80.

Treatment	No. of	Rate per		onals for (Untr. = 100)	Untreated	Per cen	t effect
	expt.	100 kg seed -	non- mercury	mer- cury	- % attack	non- mercury	mer- cury
Mercury compound							
Ceresan	15	200 g		106	33,1		99,9
Non-mercury compounds		_			,-		00,0
Baitan F-powder	3	200 g	102	115	37,3	100	100
Baitan F-liquid	2 ·	200 ml	107	109	31,2	100	100
BAS 389 01F	1	$200  \mathrm{ml}$	101	101 .	13,2	100	100
BAS 3302 F	2	200 g	111	118	54,2	100	100
	1	300 g	104	99	85,5	100	99,2
Bayer 5488	1	200 g	101	109	28,0	100	100
Bayer 6743	2	200 g	105	103	31,2	99,4	100
Bayer 6744	2	200 ml	102	103	31,2	100	100
Benlate	<b>2</b>	100 g	100	102	17,6	100	100
	1	150 g	111	113	71,1	100	100
	3	200 g	103	100	15,7	98,3	100
Benlate + Pomarsol F	1	100  g + 100  g	126	122	37,2	100	100
Benlate + Trimangol	1	100  g + 100  g	113	113	71,1	100	100
Busan 72	1	65 ml	98	96	12,8	96,1	100
KVK/Busan	2	200 ml	90	101	10,0	100	100
Derosal	4	200 g	106	107	46,3	99,6	99,6
Dithane M-45	1	200 g	93	109	28,0	100	100
Dithane Z-78	1	200 g	104	109	28,0	96,8	100
DP — carbendazim	1	300 ml	114	117	49,2	100	100
Folcidin	2	150 g	117	111	61,4	96,9	99,6
Fungaflor-liquid	1	300 ml	98	99	85,5	38,9	99,2
Granosan	3	200 g	103	101	49,3	97,8	99,4
Panoctine	2	200 ml	99	103	31,2	99,5	99,5
Panoctine Plus	3	$200  \mathrm{ml}$	101	101	22,4	100	99,3
Panocține Universal	2	200 ml	101	112	27,1	100	100
PL 3338	1	200 ml	96	97	4,7	100	100
PL 3417	1	200 ml	96	126	49,5	97,2	100
PL 3418	1	200 ml	100	126	49,5	97,2	100
Pomarsol Forte	1	200 g	105	109	28,0	100	100
Rovral	1	200 g	99	97	4,7	100	100
Scorvine	1	200 g	106	95	16,2	96,9	100
Sidipreg	3	200 ml	114	111	64,6	82,2	99,7
Sidipreg 77	1	200 ml	110	126	49,5	92,3	100
Sisthane	1	240 ml	114	126	49,5	100	100
CMTB 30 EC	1	100 ml	105	99	85,5	94,4	99,2
	3	200 ml	100	108	54,2	98,8	99,4
Topsin M	1	50 g	111	113	71,1	98,0	100
	4	200 g	105	116	35,2	95,0	100
Frimangol	1	150 g	100	109	28,0	100	100
Frimidal 10 S	3	200 ml	91	113	34,5	100	100
Vitavax	3	200 g	104	99	10,9	100	100
Vitavax T-liquid	5	300 ml	106	105	38,0	95,3	99,6
Voronit	1	200 g	103	109	28,0	100	100
Voronit-liquid	4	300 ml	112	110	58,4	99,4	99,6
-	1	500 ml	109	114	32,2	100	100
	1	1000 ml	114	114	32,2	100	100
Voronit special	$ar{2}$	200 g	100	112	27,1	100	100
'118/1A	3	200 g	97	113	34,5	100	
051/1	$\overset{\circ}{2}$	200 g	102	122	49,4	100	100
051/3A	1	200 ml	107	117	49,4		100
· <del></del>	$\overline{2}$	300 ml	96	112	40,4	100	100

Table 6. Effect of seed treatments on loose smut of oats, 1965-80.

	No. of	Rate per	Proporti emergence (	onals for $Untr. = 100$ )	Untreated	Per cent	effect
Freatment .	expt.	100 kg seed	non- mercury	mer- cury	% attack	non- mercury	mer
Mercury compound		•					OH 5
Ceresan	15	300 g		112	65,4		87.7
Non-mercury compounds							=
Baitan F-powder	2	200 g	101	103	92,9	100	76,8
Baitan F-liquid	2	200 ml	101	99	87,2	100	96,4
BAS 389 01F	1	200 ml	102	100	85,7	100	99,2
BAS 395 03F	1	200 ml	101	100	85,7	100	99,2
BAS 3302 F	1	200 g	121	151	60,9	100	88,5
D110 0001 1	2	300 g	105	105	80,5	99,5	80,1
Bayer 5488	1	300 g	111	113	51,3	15,4	97,5
Bayer 6743	2	200 g	118	115	74,3	99,7	76,1
Bayer 6744	2	200 ml	112	115	74,3	99,6	76,1
Benlate	ī	100 g	100	110	34,5	96,5	91,9
Delitate	ī	200 g	106	110	34,5	100	91,9
	$\overline{2}$	300 g	144	129	36,8	100	93,8
Benlate + Pomarsol F	ī	100 g + 100 g	161	151	60,9	100	88,5
Benlate + Trimangol	ī	100 g + 100 g		108	68,6	100	87,3
Busan 72	î	100 ml	106	110	83,2	34,7	78,8
	2	300 ml	121	129	36,8	78,8	93,8
KVK/Busan	4	200 g	121	120	75,5	99,8	78,
Derosal	1	. 300 g	106	113	51,3	24,6	97,
Dithane M-45	1	300 g	95	113	51,3	9,0	97,
Dithane Z-78	1	300 ml	99	98	88,7	95,6	93,
DP — carbendazim	$\overset{1}{2}$	150 g	123	126	76,7	98,8	80,
Folcidin		450 ml	95	101	92,4	10,1	72,
Fungaflor-liquid	1		110	110	80,3	99,4	75,
Granosan	3	200 g	88	115	74,3	9,9	76,
Panoctine	2	200 ml	77	115	74,3	5,8	76,
Panoctine Plus	2	300 ml	100	105	79,1	91,2	93,
Panoctine Universal	1	200 ml		103	97,1	90,3	60,
	1	300 ml	101		,	50,3	97,
Pomarsol Forte	1	300 g	103	113	51,3	— 1,6	93,
Rovral	1	200 g	105	105	79,1		76,
Scorvine	1	300 g	87	95	65,1	8,3	87,
Sidipreg	· 1	300 ml	107	108	68,6	31,3	
	2	400 ml	125	126	76,7	40,3	80,
Sisthane	2	200 ml	98	102	83,9	98,1	93,
TCMTB 30 EC	1	100 ml	101	101	92,4	11,1	72,
	3	$200  \mathrm{ml}$	128	125	78,9	34,1	80,
Topsin M	1	75 g	109	108	68,6	78,0	87,
-	1	200 g	136	151	60,9	54,2	88,
Trimangol	1	225 g	110	113	51,3	73,1	97,
Trimidal 10 S	3	200 ml	103	104	88,3	98,4	82,
Vitavax	3	300 g	119	<b>122</b>	52,3	100	88,
Vitavax T-liquid	5	300 ml	121	117	76,2	97,4	81,
Voronit	1	300 g	109	113	51,3	12,7	97,
Voronit-liquid	1	750 ml	90	106	48,6	18,3	96
. Ozomi mana	ī	1500 ml	98	106	48,6	47,1	96,
7118/1A	3	300 g	100	104	88,3	99,0	82
9051/1	2	200 g	103	103	92,9	95,0	76,
9051/3A	1	200 ml	103	98	88,7	97,3	93,
9001/3W	1	300 ml	104	108	97,1	88,0	60,
	1	450 ml	102	105	79,1	100	93,

Table 7. Effect of seed treatments on yield of barley and wheat.

			Leaf stri	Leaf stripe of barley	ley		Loose smt	Loose smut of barley	sy .		Loose smut of wheat	t of whe	
Treatment	Rate per 100 kg seed	No. of expt.	% attack Un- Pr treated u	ack Prod- uct	Proportionals for yield (Untr. = 100)	No. of expt.	% attack Un- Pr treated u	lack Prod- uct	Proportionals for yield (Untr. = 100)	No. of expt.	% att Un- treated	attack Prod- l uct	Proportionals for yield (Untr. = 100)
Mercury compound Ceresan Non-mercury compounds	200 g	ت د	26,6	0,2	138		:		·				
Baitan F-lowder Baitan F-liquid BAS 330? F	200 g 200 ml	Н	9,2	6,2	116	4:01	11,7 14,6	0,6 0,8	114 110	ന ന	6,1 4,7	0,4 0,2	104 99
Bayer 6743 Bayer 6744	200 g 200 g 200 ml	нп	58,4 58,4	58,8 45,2	99 115	Н	9,1	0,4	94		0, 1, 1, 0, 6, 6,	0,0 7,0	113 92
Benlate + Pomarsol F Benlate + Trimangol	$50\mathrm{g} + 100\mathrm{g} \ 100\mathrm{g} + 100\mathrm{g} \ 50\mathrm{g} + 100\mathrm{g}$						9,1	6,3 6,3	106 85	·	- 4, 4, 6 5, 4, 6, 6	9,00	96 94
Derosal	$100  \mathrm{g} + 100  \mathrm{g}$ $200  \mathrm{g}$	-	58,4	64.4	75	·	, 4, 0 , 2, 1	0,8 7	89 89	H نا د	7, 0, <del>4</del>	0,4 6,4	83 102
Fungaflor-powder Fungaflor-licuid	300 g 300 m	<b>63</b> 0	16,2	1,2	112	1	160	- 5	r S	4	ή, δ	۲,6	5 8
Granosan	200 g	, 	58,4	35,7	137					,	7.3	6	04
Panoctine Panoctine Plus	200  ml 200  ml	<b>⊣</b> ≈1	58,4 40,4	52,1	106 145					l <del>eri</del> +	7,3	, 7. I	101
Panoctine Universal	200 ml	<b>6</b> 7 -	16,2	0,3	114	က	8,3	2,5	107	⊣ ന	2,7 7,6	3,1	101
Sidipreg	200 g 200 ml	7	22,3	3,0	114		7,5 9,1	7,8 5,6	88 88	-1 6	6,5 2,2	7,0	96
Sisthane	$\frac{400 \text{ ml}}{200 \text{ ml}}$	63	15,8	0,1	118	_ ;	4,2 16,1	2,1	97 107	ı ຄາ	į r	î -	, ( <u>)</u>
Topsin M		1	10,1	0	115	27	8,8 4,2	0,1 2,2	113	. ca –	. 80 c	0,0	109
:	.200 g . 250 g					. 4	, 16	, e	64		2,2,	2,1	85
Trimidal 10S	200 ml	ლ –	13,9 33,1	5,6	) 110 101	4-	10,3	0,4	105	ري ري	6,7	9,0	86
Vitavax		ı	1(2)	2	001	- <del>-</del>	<ul><li>19,1</li><li>0,1</li></ul>	, 0,7	100	- 2	0,8 0,2	< 0,1 < 0,1	17 102
Vitavax T-liquid		П (	22,3	3,5	103	മയ	8,4 11,5	0,1 0,8	104 100	11 9	4,5 0,0	0,5	101 102
9051/1	200 g 200 g	70 cz	13,9 9,7	0,0	119 118	4 4	10,3 11,7	6, c 6, c	102	លេយ	6,7	, 67 c	103
9051/3A	200 ml	6	9,2	0,6	120	67 6	14,6	, e	105	ကြေ	4,7	2,2	102 106
		3	10,2	1,1	CII	2	8,3	8,0	110	က	2,6	1,8	102

the leaf stripe control tests, both with mercury dressing and with almost all non-mercury dressings, substantial yield increases were obtained. In most cases the yield increase was greater than would be expected from a mere decrease in leaf stripe disease. In the tests on loose smut of barley and loose smut of wheat, the situation was just about the reverse. The yield increases obtained with the seed dressing were usually small, only rarely matching the reduction in the percentage of smut.

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#### SELOSTUS

# Kevätviljojen peittauskokeita elohopeattomilla valmisteilla

#### REIJO VANHANEN

#### Maatalouden tutkimuskeskus

Kasvitautiosastolla on vuosina 1965—80 tutkittu usean kymmenen elohopeattoman peittausaineen tehoa ja käyttökelpoisuutta kevätviljojen siemenlevintäisten tautien torjunnassa. Tavoitteena on ollut löytää elohopeapeittausta myrkyttömämpi ja ympäristölle vaarattomampi vaihtoehto. Kenttäja kasvihuonekokeissa on selvitetty valmisteiden tehoa ohran viirutautia, ohran ja vehnän lentonokea, vehnän haisunokea sekä kauran avonokea vastaan. Lisäksi on tutkittu aineiden vaikutusta viljan orastumiseen ja satoon. Viirutauti- ja lentonokikokeissa on käytetty luontaisesti saastuneita siemeniä, kun taas avo- ja halsunokikokeissa siemenet on saastutettu keinollisesti noki-itiöillä.

Lähes kaikki kokeillut peittausaineet tehosivat erinomaisesti vehnän haisunokeen. Useimmilla valmisteilla saatiin hyvä tulos myös kauran avonoen torjunnassa. Sitä vastoin lentonokea ja viirutautia vastaan vain harvat aineet olivat tehokkaita. Suurin osa elohopeattomista valmisteista paransi hieman viljan orastuvuutta. Viirutaudin torjuntakokeissa peittauksella saatiin huomatta-

via sadonlisäyksiä, mutta lentonokikokeissa satoa lisäsi ainoastaan muutama valmiste.

Laajatehoisten elohopeattomien peittausaineiden löytäminen on osoittautunut vaikeaksi. Joidenkin valmisteiden karsiutumisen syynä on ollut niiden myrkyllisyys joko ihmisille tai kasveille. Valtaosa elohopeattomista yhdisteistä on kuitenkin jouduttu hylkäämään tehon rajoittuneisuuden vuoksi. Sisävaikutteisten tehoaineiden kehittäminen ja eri aineiden yhdistäminen on laajentanut elohopeattomien valmisteiden vaikutusaluetta, vaikkakaan toistaiseksi ei vielä ole saatu käyttöön yhtään sekä nokitauteihin että viirutautiin hyvin tehoavaa peittausainetta.

Neljällä elohopeattomalla valmisteella on nykyisin Suomessa myyntilupa kevätviljojen tautien torjuntaan, nimittäin Benlatella kauran avonokea ja vehnän haisunokea vastaan, Panoctine Plus-valmisteella viirutautia ja haisunokea vastaan sekä Vitavaxilla ja Vitavax T-nesteellä lentonokea, haisunokea ja avonokea vastaan.

# Seria PHYTOPATHOLOGIA N. 78 — Sarja KASVITAUDIT n:o 78

# WINTER DAMAGE AND LOW-TEMPERATURE FUNGI ON LEYS IN NORTH FINLAND IN 1976—1979

#### KAIHO MÄKELÄ

MÄKELÄ, K. Winter damage and low-temperature fungi on leys in North Finland. Ann. Agric. Fenn. 20: 102—131. (Agric. Res. Centre, Inst. Pl. Path. SF-01300 Vantaa 30, Finland).

The material comprised a total of 740 fields in 11 communes. The samples were collected springs and studied in the laboratory microscopically.

The three-fourths of the leys, consisted of timothy, whereas timothy-meadow fescue and timothy-Poa alpigena (Fr.) Lindm. mixed leys comprised 10  $^{0}$ / $_{0}$  each. Three-fourths of the leys were on mineral soil and one-fourth on peat soil.

The spring of 1977 had the most winter-damaged leys (57  $^{0}/_{0}$ ), while 1979 had the fewest (28  $^{0}/_{0}$ ).

The most common low-temperature fungi were Sclerotinia borealis (Bubák and Vleugel), Typhula spp. and Fusarium spp. These fungi together were more common in the eastern and northern parts of Lapland than in the western and southern parts. The annual variations in abundance of these fungi corresponded with the variations in winter damage.

S. borealis was found in an average of 71  $^{0}/_{0}$  of the leys. It was most common and abundant in first-year timothy leys. Also in recently cleared, newly established land, fungal injuries could be found already in the first year. No consistent differences were observed in leys established at different times or by different methods, nor between leys located on mineral or peat soils.

The sclerotia of *Sclerotinia borealis* were found within the plant matter, almost entirely on the surface of the ground or in the uppermost 1,5 cm layer. The sclerotia remained viable on the ground surface at least two years. Similarly, apothecia developed only in sclerotia lying on the soil surface.

Typhula ishikariensis Imai occurred in the whole experimental material, averaging  $55\,^{0}/_{0}$ . It caused damage principally in older timothy leys. Typhula incarnata Lasch ex Fr. appeared in only  $10\,^{0}/_{0}$  of the experimental material and was thus relatively unimportant.

Fusarium species occurred in an average of  $44\,^{\circ}/_{\circ}$  of the investigated leys. In timothy their significance was minor than meadow fescue. The most common Fusarium species were F. nivale (Fr.) Ces., F. avenaceum (Corda ex Fr.) Sacc., F. culmorum (W. G. Smith) Sacc. and F. semitectum Berk. & Rav.

Large numbers of other fungi were also determined in the leys of this investigation, totalling about 75 species belonging to 64 genera.

Index words: Low-temperature fungi, winter damage, leys, Phleum pratense, Sclerotinia borealis, Typhula species, Fusarium species.

#### INTRODUCTION

Grasslands have a greater significance in North Finland (ca. 65-70°N, 21-30°E) than in the rest of the country. In the years 1975—1978 about 70 % of the cultivated land was leys while the corresponding figure in the whole country was only 36%. The proportion of hay leys (Fig. 1) was greater (68 %) than the average for the country (58 %), silage leys (15 %) were about the same as for the whole country (18%), while pasture (17 %) was less than for the country as a whole (24 %). The average yield of ley grass in North Finland (3370 kg/ha) was somewhat smaller than that for the whole country (3790 kg/ha) (Off. Statist. Finl. Agric. 1975, 1976, 1977, 1978).

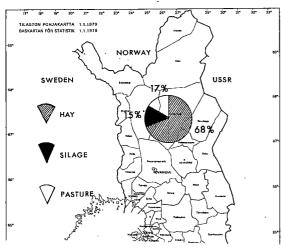


Fig. 1. Location of leys in Lapland province, 1975—1978.

The most important cultivated grass and the major ley plant is timothy (Phleum pratense L.) (Hakkola 1980). In addition, meadow fescue (Festuca pratensis Huds.) is grown to some extent, principally in mixtures with timothy. As a green fodder crop, annual ryegrass (Lolium multiflorum, Lam.) is cultivated (Raininko 1976). Of the wild grasses Poa alpigena (Fr.) Lindm. is prevalent and valuable. It occurs throughout all of Lapland, and the further north one goes, the more common it becomes (Roivainen 1937, Valmari 1979). Other wild fodder grasses worth mentioning are brown top bent (Agrostis tenuis Sibth.), red fescue (Festuca rubra L.), reed canary grass (Phalaris arundinaceae L.) (Roivainen 1937), meadow foxtail (Alopecurus pratensis L.) (Pohjakallio and Salonen 1956), as well as couch grass (Agropyron repens (L.) PB) (Teräsvuori 1933).

Winter damage to leys in North Finland can be severe in certain years (Jamalainen 1967, 1970, Pohjonen 1976). For instance, at the experimental station at Apukka (66°35'N, 26°00'E) during the 27-year period 1951—1977 timothy overwintered poorly in 12 years, while meadow fescue overwintered poorly in 9 years during a 26-year period (Jamalainen 1978). At the Muddusniemi Experimental farm (69°05'N, 27°03'E) during the 15-year period 1950—1965 timothy showed poor winter survival in 6 years while meadow fescue during the same period overwintered well or satisfactorily (Nissinen and Salonen 1972 a).

In earlier years timothy was considered to be quite resistant to winter damage in Lapland (Isotalo 1959). At the time when grass fields were used mainly for producing hay and were given moderate fertilization, timothy leys persisted for long periods, 6-8 years (Roivainen 1937, Aikkinen 1951). The same observation has been made by numer-In recent decades, however, ous farmers. with increased emphasis on animal husbandry and leys being cultivated intensively to produce silage (»green line» cultivation), unexpected problems with overwintering have arisen (Pohjonen 1976, Valmari 1979). According to farmers, grass fields at the present time last 3-4 years at the most, often only 1-2 years. In Lapland leys nowadays must be ploughed up at a young age (Pohjonen 1976), often after only one year (Valmari 1979). Each year about 12 500 hectares of grassland are renewed, which amounts to one-fourth of the ley area in North Finland (Maaseudun Tulevaisuus 1980, no. 67).

The poor overwintering of leys depends on many different factors, such as plant species and variety, soil condition and fertilization, sowing and cutting time, fungal damage, etc. (Pohjonen 1976, Arsvoll 1977, Valmari 1979). Other factors contributing to decrease in yields are too small field area in relation to the number of animals, continuous grass cultivation on the same field, and heavy farm machines (Mäkelä 1980). In addition, other causes may be poor drainage, weeds, improper pasturing methods, and above all, deficiencies and incorrect proportions of plant nutrients (Marjanen et al. 1979). Overwintering of leys is also influenced by the weather conditions in the winter and spring as well as by all the cultivation procedures in the previous growing season from the establishment of the ley to the last cutting (Pulli 1976, 1980).

Winter injuries are also due to unfavour-

able physical conditions as a result of the weather. These so-called abiotic factors include frost, ground heaving, ice and water cover (Ylimäki 1955, Jamalainen 1956, 1978, Blomqvist 1970, Arsvall 1973, Andersen 1980). Such winter injuries are less prevalent in North Finland than in other parts of the country (Blomqvist 1970, Jamalainen 1978). The thick snow cover in North Finland which lasts from November to May protects the plants from freezing. The occurrence of freezing injuries depends primarily upon how early in the spring the snow melts (Pohjakallio and Salonen 1956, Isotalo and Vogel 1962, Vuorinen 1979). Plant damage due to standing water occurs in North Finland mainly on peat soils (Jamalainen 1970, Valmari 1979). Timothy is resistant to water cover for as much as three weeks, depending on the conditions. Similarly, timothy is more resistant to a high level of ground water than other ley grasses (Saukko 1946).

The most important cause of winter damage in the areas of northern Finland with a heavy snow cover are considered to be biotic factors. The most severe damage has occurred in years when after a long, wet autumn snow fell on unfrozen ground, and a thick snow cover lasted for a long time and melted slowly (Jamalainen 1949, Ekstrand 1955, Nissinen and Salonen 1972 a, Årsvoll 1973). In contrast, freezing of the ground before the fall of snow effectively prevented damage (Isotalo and Vogel 1962).

The principal cause of poor overwintering of leys in Lapland is generally held to be low-temperature parasitic fungi: Sclerotinia borealis Bubák and Vleugel, Typhula spp. and snow mould (Fusarium spp.) (Jamalainen 1970, 1978, Pohjakallio and Salonen 1956, Nissinen and Salonen 1972 a). In addition to these, there are a large number of other fungi which attack overwintering grasses, at least under Norwegian conditions (Arsvoll 1975).

This present investigation was started as a result of widespread damage to overwintering leys in North Finland in the early years of the 1970's. In order to study this problem, the Agricultural Research Centre carried out extensive investigations which were partly financed by the Department of Forestry and Agriculture through project no. 12—3 dealing with north-Finnish ley damage. These studies were performed jointly by the local field trial agency, the departments of Plant Breeding, Plant Pathology, and Agricultural Chemistry and Physics, as well as the experimental stations of Carelia, North

Savo, Central and North Pohjanmaa, and Lapland. In addition the Kainuu and Lapland Agricultural Centres participated mainly within the framework of their own plans.

The Department of Plant Pathology carried out investigations in the years 1976—1979 with the aim of determining the parasitic fungi occurring in leys in North Finland, their incidence in different years, different areas and different kinds of fields. Emphasis was given to the species of fungi, and their relative amounts and prevalence. In addition, other fungal species found in the grass samples were also identified.

#### MATERIAL AND METHODS

The experimental material consisted of grass samples (Table 1) collected in the spring during the end of May and the beginning of June soon after the snow had melted. The dates of collection were: 20.—29. 5. 1976,

24. 5.—7. 6. 1977, 29. 5.—8. 6. 1978, 5.—15. 6. 1979. The samples were taken principally from leys which had suffered winter damage and which were located on the fields of individual farmers, agricultural schools and

Table 1. Numbers of farms and leys investigated, 1976—1979, by localities.

					Num	pers of sa	amples in	vestigate	đ		
		19	976	1	977	1	978	1	979	1976-	<b>—1979</b>
Localities	Biological provinces 1	Farms	Leys	Farms	Leys	Farms	Leys	Farms	Leys	Farms No. per year	Total No.
Tornio	PP	5	9	2	6	9	11	8	8 .	6,0	. 34
Ylitornio	) <b>PP</b>	5	10	3	4	7	. 9	6	9	5,3	32
Tervola	PP ·	9	15	13	19	18	31	12	28	13,0	93
Rovaniemi	PP	8	34	12	35	1.8	32	14	35	13,0	136
Kemijärvi	$_{\mathrm{PP}}$		_	4	9	10	14	13	15	9,0	38
Salla	Ks .	4	12	4	12	3	12	5	7	4,0	43
Pelkosenniemi	KemL			3	3	4	·6	8	8	5,0	17
Sodankylä	KemL	10	17	21	35	22	42	31	41	21,0	135
Kittilä	KemL	11	38	9	23	20	33	18	22	14,5	116
Muonio	KemL	_	_	4	7	5	20	5	13	4,7	40
Inari	InL	6	10	4	8	5	19	4	20	4,8	57
Total		58	145	79	161	121	229	124	206		741

<sup>1</sup> PP = North Ostrobothnia

Ks = Kuusamo KemL = Kemi Lapland

KemL = Kemi Lapland InL = Inari Lapland

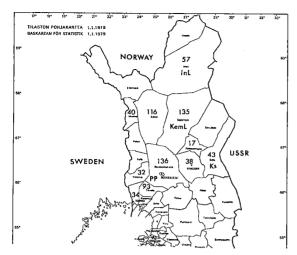


Fig. 2. Total number of leys investigated, by localities, 1976—1979.

the Lapland experimental station. In the four years 1976-1979 a total of 740 fields (varying 145-229 in the different years) in 11 communes were investigated (Fig. 2). In addition, the farmers were interviewed with respect to questions involving ley cultivation. In this way useful supplementary information was obtained. The severity of damage to the ley was estimated visually at the site, using a 6-point scale, in which the values were: +3 very good, +2 good, +1 moderately good, —1 moderately poor, —2 poor, —3 very poor. At the same time the abundance of the three most important low-temperature parasitic fungi were estimated (S. borealis. Typhula spp., Fusarium spp.). The following 4-point scale was used: 0 = fungus not found, 1 = small amounts found, 2 = moderate amounts, 3 = abundant amounts found.

Samples were taken from the leys and detailed studies were later made in the laboratory on the sclerotia occurring in the samples. The fungi were cultured in Petri dishes using the moist chamber method, keeping the dishes at room temperature and alternately at  $\pm 10^{\circ}$ C for about one month. The fungi were investigated microscopically.

The occurrence of sclerotia of Sclerotinia borealis at different depths in the soil was studied in the spring of 1978 and 1979. In leys which were severely infested with the fungus, pieces of sod ( $10 \times 10 \times 10$  cm) were removed and subsequently examined in the laboratory for the numbers and position of the sclerotia.

In the years 1978—1980 studies were made on the preservation of sclerotia and the development of apothecia of S. borealis under natural conditions. These studies were made at various trial locations both on the ground surface and at a depth of 5 cm. The sclerotia were preserved in tight-meshed metal net bags (10 imes 10 cm) each containing 100 sclerotia. These were examined at intervals of 1/2-1 year, one time after the snow had melted in the spring and the other time before the fall of snow in the autumn. There were five trial locations: Muddusniemi experimental farm at Inari (69°04'N, 27°03'E), the school of agriculture at Kittilä (67°41'N, 24°54'E), the Lapland experimental station at Rovaniemi (66°35'N, 26°00'E), the North Savo experimental station (63°09'N, 27°19'E) and the Department of Plant pathology at Tikkurila (60°17'N, 25°04'E).

The occurrence of apothecia of S. borealis in different leys was investigated just before the arrival of snow, 13.—16. 10. 1980. Observations were made at 30 leys in the region comprising Sodankylä, Rovaniemi and Tervola. Most of the leys were young, first-and second-year timothy leys, three-fourths of them were on mineral soil and one-fourth on peat soil. The numbers of apothecia were counted on a trial plot  $15 \times 15$  cm (225 cm²). There were a total of 125 such plots distributed among 30 leys.

On these same leys observations were made on the development of sporophores of *Typhula ishikariensis*.

The growth of S. borealis on leys under

the snow in late winter and early spring was studied at the Lapland experimental station in the years 1977—1979. Continuous temperature measurements were made at the ground surface under the snow. Samples were taken from the leys at approximately two-week intervals from the end of March until the beginning of May. The fungi growing in these samples were subsequently studied.

#### Weather Conditions

In the area of this study in North Finland the thermal winter  $(0^{\circ}-0^{\circ}C)$  is long, averaging 175—200 days. The thermal growing season  $(5^{\circ}-5^{\circ}C)$  is correspondingly short, averaging 115—140 days. The growing season usually begins in the latter half of May (10.—25. 5.) and terminates at the end of September (20.—30. 9.). Autumn and especially spring are short (Kolkki 1966).

The snow cover arrives early, already in October and it does not disappear until May. Although the depth of the snow cover varies in different years and in the different parts of the region, from 30-40 cm in the northernmost parts to 70-80 cm in the central and southeast parts (Huovila 1970), it is heavy enough in the whole region to protect the leys from even the most severe freezing temperatures (Vuorinen 1979). A layer of snow as little as 15-20 cm succeeds in keeping the ground surface temperature above — 5°C when the air temperature is  $-16^{\circ}$  to  $-30^{\circ}$ C. When the snow cover is at least 25 cm thick the ground surface remains above -2°C even at air temperature of -30°C (Ylimäki 1962). In the years of this study the winters were colder than normal. In the winter 1976 -77 the depth of snow was exceptionally great. On the other hand, the winter of 1978 -79 had less snow than usual (Fig. 3). In other years the thickness of the snow cover

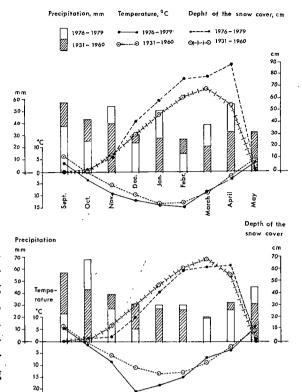


Fig. 3. Weather conditions in the winter seasons 1976-1977 and 1978-1979 at the Sodankylä observatory (67°22'N, 26°39'E).

was approximately normal (Meteorol. Yearb. Finl. 1975, 1976, 1977, 1978, 1979).

Winter 1975—76. The first snow fell in North Finland 30. 9.—16. 10. The lasting snow cover finally came 16. 11., 2—3 weeks later than usual. January was much colder than normal. February and March were mild. The latter half of April was cold. The snow cover disappeared about a week or two earlier than usual, on 10.5. in the areas around Apukka and Salla, and on 15. 5. in the Sodankylä and Inari region. The latter half of May was exceptionally warm.

Winter 1976—77. In most of North Finland the lasting snow cover came already at the end of September, 3—4 weeks earlier than normal. During the whole winter of 1976—77

the snow cover was considerably deeper than normal, especially towards the end of the winter. The temperature was much below normal, already starting in September, but especially in November and December. January likewise was cold and February even colder. In the early part of March the temperature rose considerably above normal, but at the end of the month it dropped sharply below normal. April was cold with abundant snowfalls. In May the weather was very warm. The snow cover disappeared at the usual time causing profuse melting water and floods which occurred around 15.5. in the Apukka and Salla region and 20.5. in the Sodankylä and Ivalo region.

Winter 1977—78. The first snow came 12.—20. 9., or about three weeks earlier than the average. The lasting snow cover came about two weeks earlier than normal. Before this the ground froze deeper than usual. The entire winter season, including the autumn,

was colder than normal. Especially in February there were low temperatures. The snow depth was generally normal. In the spring the snow disappeared 13.5. at Apukka, 18.5. at Salla and Sodankylä, and 24.5. at Ivalo. At the end of May it was very hot.

Winter 1978-79. The ground froze in the first part of October. The lasting snow cover did not come until mid-November, which was 2-3 weeks later than normal. During the whole winter the snow depth was exceptionally small. The temperatures during November, December, January and February remained below normal. Especially December and January were extremely cold. The first part of March was unusually warm, while the last part became very cold. April and the beginning of May were colder than normal. On May 14 snow fell in North Lapland. Warm weather did not prevail until 18.5., and the snow cover disappeared slightly later than usual.

#### RESULTS AND DISCUSSION

#### Grass species

The majority of the leys investigated were timothy leys (Fig. 4): 90 % in the years 1976 and 1977, 55 % in 1978 and about 60 % in 1979. The north Finnish pasture grass mixture (timothy 55—60 %, meadow fescue 40—45 % comprised only a few % in the first two years and 10—14 % in the last two years. The Poa alpigena-timothy grass mixture comprised nearly 20 % in 1978 and 1979, while in the same years pure Poa alpigena leys made up about 10 %.

According to information from farmers, grass fields established on peat soils are almost always pure timothy leys. Also on mineral soils pure timothy leys are predominant. In addition to timothy leys, pasture grass mixtures are also grown in North Finland.

Many farmers have observed that such mixtures are more suitable for pasturing and more palatable to cattle than pure timothy. Similarly such mixtures are considered to provide better fodder for sheep and reindeer than pure timothy, especially older, coarser stands. Some growers find it necessary to use the north Finnish pasture grass mixture because of a lack of timothy seed. Some farmers do not approve of the pasture mixture because they believe that it is less dependable than leys consisting of pure timothy.

Poa alpigena, which is found everywhere in Lapland as a wild grass, appears naturally in cultivated leys and can also grow as pure stands. It has long been an important fodder crop in Lapland and even today it provides part of the fodder for farm animals in years of severe winter fungal damage (Roivainen

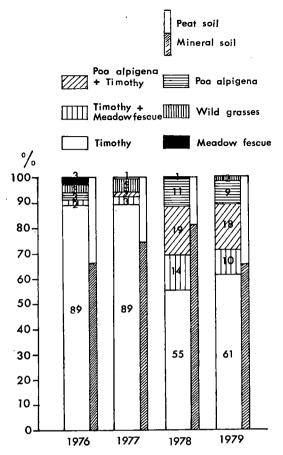


Fig. 4. Plant species and soil types on the leys investigated, 1976—1979.

1937, Valmari 1979). According to Valmari (1979) Poa alpigena is suitable especially as a silage and pasture crop.

### Soil

The majority of the leys investigated (av. 72%, range 65—84% in the different years) were on mineral soils (Fig. 4). The reason for this is that, according to the opinion of the farmers, leys in North Finland have poorer overwintering on mineral soils than on peat soils. On the other hand, the proportion of leys on peat soils is very large,

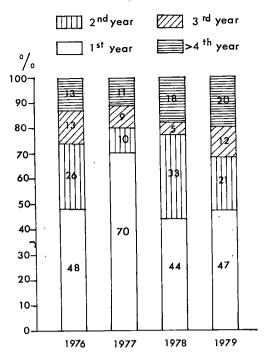


Fig. 5. Age of leys investigated, 1976-1979.

amounting to nearly one-half (Isotalo 1959, Elonen 1976). In 1951 46 % of the newly established leys in North Finland were located on peat soils (Paatela 1953). According to Årsvoll (1973) abiotic factors and S. borealis cause considerably greater damage to leys on peat than on mineral soil.

# Age of ley

Most of the leys studied were young (Fig. 5). First-year leys made up an average of 52 % (annual variation 44—70 %), second-year leys comprised 23 % (variation 10—33 %), third-year leys were only 10 % (range 5—13 %), and fourth-year or older leys averaged 15 % (range 13—20 %).

First-year timothy leys were chosen in greater numbers for this study than older

leys for the reason that they have been shown by numerous investigators to be more susceptible to winter damage than older leys (Pohjakallio and Salonen 1958, Nissinen and Salonen 1972 a, Jamalainen 1970). Many leys, owing to such damage, must be partially resown already after the first winter. Nowadays most leys are short-lived (Pohjonen 1976, Valmari 1979). In older leys there are abundant wild grasses and weeds which hinder the observations.

### Time of ley establishment

The majority of the leys investigated (av. 75 %, annual range 72—86 % were established at the beginning of summer (Fig. 6). It has been found in many studies (Nissinen and Salonen 1972 b. Valmari 1979) that early sowing produces a more winter-resistant lev and larger yields. A small number of the leys were sown at the end of summer (August), and the shoots of these plants remained small and delicate. Such shoots have been found to be susceptible to winter fungi (Nissinen and Salonen 1972 b) and the yield in the following summer was reduced. Grasses which are sown in the autumn, in September and October, do not germinate until the following spring. Such leys increase in number as one goes towards the north. Different sowing times are generally used on the same farm in order to equalize the yields and to ensure a successful establishment of the ley.

#### Companion crop

About one-half of the leys investigated were established without a companion crop (Fig. 6). Of the companion crops barley was the most common, making up about one-fourth of the leys, oats were used in  $13 \, {}^{0}$ /o of the leys, and rape  $6 \, {}^{0}$ /o. Other crops occasionally used were

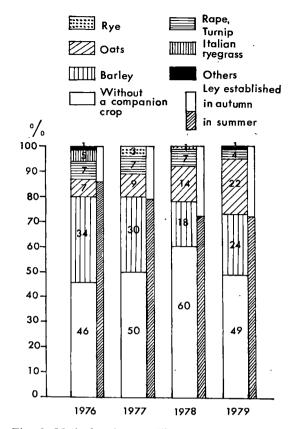


Fig. 6. Methods of ley estiblishment, 1976—1979.

rye, Italian ryegrass and turnip. Fallow was virtually not employed at all. According to studies made in 1951 (Paatela 1953) the plants used as companion crops on leys in Lapland were mainly barley and green fodder on mineral soils and oats, barley and green fodder on peat soils. According to other workers (Nissinen and Salonen 1972 b, Valmari 1979) leys can be more successfully established without a companion crop. The use of a companion crop did not have an influence in all of the trials upon the overwintering of firstyear leys (Pohjakallio and Salonen 1956). In many other cases, however, companion crops have ensured the succes of ley establishment (Isotalo 1960, Nissinen and Salonen 1972b).

At the present time there is great varia-

Weeds

tion among farmers in the use of a companion crop. The method without a companion crop is employed for almost all levs on peat soils as well as those established in the late summer and autumn on mineral soils. Also some of the leys established in the spring on mineral soils are sown without a companion crop. Companion crops are used generally in the southern parts of the region. Barley was the main companion crop previously, but it is being replaced nowadays by oats. In going towards the north, the use of companion crops decreases and oats becomes the dominant plant used. Especially in the southern areas there have been fields where lodged barley caused bare patches in the ley. Attempts have made to reduce the unfavourable effects of the companion crop stubble as well as the dead foliage of the old ley by burning over the field in the early spring. This practice is particularly common in the southern parts of the region. Too early pasturing has also been known to cause bad damage to some of the first-year leys.

### Preceding crop

On the farms investigated the preceding crop before ley establishment was almost always cultivated lev. This was true on both soil types but especially on peat soils. Only in the southern areas of the region the preceding crop was to some extent barley. In a few cases the preceding crop was a green fodder crop such as pea-oats mixture, Italian ryegrass-oats mixture, Italian ryegrass, rape or turnip. Some of the leys were established on newly cleared land, both on peat and mineral soils. According to Paatela (1953) cultivated ley was the most important crop preceding the establishment of ley. In addition, other important preceding crops were barley on mineral soil and oats on peat soil. Newly cultivated lands were also quite common.

The weeds occurring in the levs were studied especially in the years 1978-1979 (Table 2). In about 15 % of the levs they occurred abundantly. On mineral soils there were more weed species (about 45) and also their numbers were greater than on peat soils. The most common weeds were: annual meadow grass (Poa annua L.), tussock grass (Deschampsia caespitosa (L) PB), Ranunculus repens L., Rumex spp., Stellaria media (L.) Vill., Taraxacum vulgaria DT, Achillea millefolium L. and couch-grass (Agropyron repens (L.) PB). In the studies of Paatela (1953 a) Poa annua was not mentioned at all and Stellaria media occurred rarely, in only 2 % of the levs. As regards the other most

Table 2. Most common weeds in the leys investigated in North Finland, spring 1978 and 1979.

Weeds			weeds no.% vestigated
Weeds	1978	1979	1978—1979
Poa annua L.	35	16	25,5
Deschampsia caespitosa			
(L.) PB.	25	24	24,5
Stellaria media (L.) Vill.	23	21	22
Ranunculus spp.	2	33	17,5
R. repens L.	31	5	18
Taraxacum vulgare DT	17	19	18
Achillea millefolium L.	14	17	15,5
Rumex spp.	11	10	10,5
R. domesticus Hartm.	8	12	10
R. acetosa L.	8	2	5
Agropyron repens (L.) PB	10	5	7,5
Cirsium spp.	5	8	6,5
Trifolium repens L.	6	7	6,5
Melandrium rubrum			
(Weig.) Garcke	5	3	4
Equisetum spp.	2	5	3,5
Barbarea vulgaris R. BR	3	3	3
Epilobium angustifolium L.	4	2	3
Rorippa islandica			
(Oeder) Borb.	4	2	3
Chenopodium album L.	3	2	2,5
Galeopsis spp.	2	3	2,5
Plantago spp.	0	5	2,5
Polygonum spp.	3	2	2,5
Trollius europaeus L.	0	4	2
Achillea ptarmica L.	3	0	1,5
Veronica longifolia L.	0	3	1,5

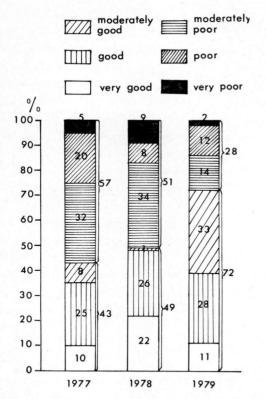


Fig. 7. Overwintering, % of leys investigated, 1977 —1979.

usual weed species, their frequency has remained quite similar.

Weeds were a serious nuisance especially in new leys which were established without a companion crop. In particular *Poa annua* appeared to be tough and resistant to trampling. It has steadily spread especially on pastured leys, and low-temperature fungithrive well in it. Weed control is not carried out to any great extent primarily because of lack of equipment.

# Winter damage

Considering the entire region as a whole, there were no years during the present study with extremely severe overwintering damage. Nevertheless, winter injury varied from year to year (Fig. 7). The greatest numbers of leys with poor winter damage were found in the spring 1977 (totalling  $57\,^{\circ}/_{\circ}$ ), and the least number ( $28\,^{\circ}/_{\circ}$ ) in 1979. Leys which had good overwintering amounted to about one-fourth of the total, and this figure was about the same in all the years. Leys with very good overwintering comprised  $10-20\,^{\circ}/_{\circ}$  and those with very poor overwintering about  $5-10\,^{\circ}/_{\circ}$ .

# Fungi causing winter damage

The most common fungi to cause winter damage in this study were *Sclerotinia borealis*, speckled snow mould (*Typhula* spp.) and snow mould (*Fusarium* spp.). They occurred, on the average, in the above order (Table 3, Fig. 8). In the whole experimental material these fungi made up an average of  $180\,^{\circ}/_{\circ}$  (variation from place to place  $148-197\,^{\circ}/_{\circ}$ ). In the eastern and northern areas of the region the fungi were more prevalent than in the western and southern areas.

There were clear annual fluctuations in prevalence of these fungi which corresponded closely to the amount of winter damage (Fig. 7). In the spring of 1976 and 1977 the fungi were more common in the leys (av.  $195~^{0}/_{0}$  and  $230~^{0}/_{0}$ ) than in the spring of 1978 and 1979 (av.  $158~^{0}/_{0}$  and  $138~^{0}/_{0}$ ).

# Sclerotinia borealis

Sclerotinia borealis is found in many grass species in the central and northern parts of Finland, in the provinces of Lapland, Oulu, Kuopio and Mikkeli (Jamalainen 1949). In Sweden the fungus occurs in the northern areas of the country as far as Dal-river (Ekstrand 1961). Also in Norway the fungus is limited to the northern regions of the country or to high altitudes in the southern re-

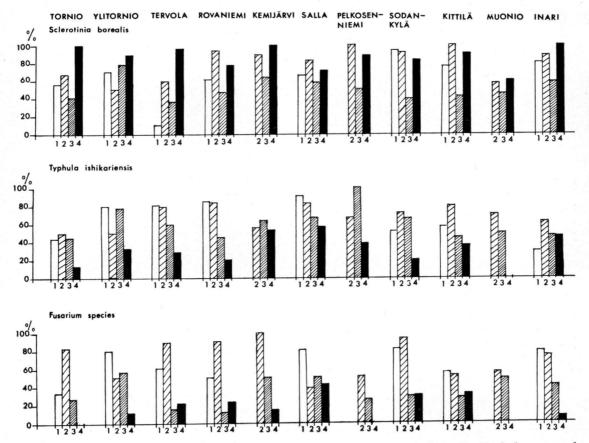


Fig. 8. Frequency of the most important low-temperature fungi, *Sclerotinia borealis*, *Typhula* spp. and *Fusarium* spp.,  $^{0}/_{0}$  of leys investigated, 1976—1979, at the different localities.

gions (Årsvoll 1975). Great damage is caused by the fungus in areas where the snow cover persists for more than 180 days or where there are more than 110—120 days with a maximum air temperature of below 0°C (Årsvoll 1973, 1975).

In the present study S. borealis occurred commonly throughout all of Lapland, in an average of 71% of the fields investigated (Table 3, Fig. 8). There were variations between the areas, the years, and especially between the different fields.

S. borealis was more prevalent in the northern areas (Sodankylä, Kittilä, Inari) where it was found in nearly 80 % of the

samples, than in the southern areas (Tornio, Tervola, Rovaniemi, Salla) where it occurred in an average of 65 % of the fields studied.

The differences between the four years were considerable. In the springs of 1976 and 1978 the fungus occurred much more rarely, in an average of 64 % and 51 % of the fields, than in the springs of 1977 and 1979 when it was found in 80 % and 88 % of the samples. The abundance of the fungus in these cases does not alone indicate the amount of damage to the leys. It is true that in the spring 1977 there were twice as many poorly-overwintered leys than in 1979. On the other hand, there were more sclerotia of

Table 3. Occurrence of the most important low-temperature fungi,  $^{0}/_{0}$  of the leys investigated in North Finland, spring 1976—1979.

			1976						19	77		
Localities	No. of samples investigated	S. borealis	T. ishikariensis	T. incarnata	Fusarium spp.	Fungi, total no%	No. of samples investigated.	S. borealis	T. ishikariensis	T. incarnata	Fusarium spp.	Fungi, total no%
Tornio	9	55	44	0	33	132	6	67	50	0	83	200
Ylitornio	10	70	80	0	60	210	4	50	50	0	50	150
Tervola	15	12	81	0	60	153	19	58	79	0	89	226
Rovaniemi	34	61	85	5	50	201	35	94	84	0	90	268
Kemijärvi <sup>1</sup>	-	_					9	89	56	0	100	245
Salla	12	66	91	0	80	237	12	83	83	25	38	229
Pelkosenniemi <sup>1</sup>	_	_			_	_	3	100	67	33	0	200
Sodankylä	17	94	52	0	82	228	35	92	72	0	94	258
Kittilä	38	76	57	7	57	197	23	100	80	13	53	246
Muonio <sup>1</sup>		_		_	_	_	7	57	71	57	57	242
Inari	10	80	30	10	80	200	8	88	63	38	75	264
Total	145						161					
11 localities, mean		64	65	3	63	195		80	69	15	66	230
1 8 » » 1 in 1977—1979	145						142	79	70	10	72	231

			1978						1979						1976—19	79	
No. of samples investigated	S. borealis	T. ishikariensis	T. incarnata	Fusarium spp.	Fungi, total no%	No. of samples investigated	S. borealis	T. ishikariensis	T. incarnata	Fusarium spp.	Fungi, total no%	No. of samples investigated	S. borealis	T. ishikariensis	T. incarnata	Fusarium spp.	Fungi, total no%
11	45	45	27	27	144	206	100	13	0	0	113	34	67	38	7	36	148
9	78	78	0	56	212		89	- 0	33	11	133	32	72	52	8	44	176
31	36	59	8	15	118	170	96	29	0	21	146	93	51	62	2	46	161
32	47	45	0	11	103	41	77	20	11	23	131	136	70	59	4	44	177
14	64	64	0	50	178	22	100	53	0	14	167	38	84	58	0	54	196
12	58	67	0	50	175	13	71	57	14	0	142	43	70	75	10	42	197
6	50	100	17	50	217	20	88	38	13	25	164	17	79	68	21	25	193
42	39	66	9	30	144	15	83	20	5	31	139	135	77	53	4	59	193
33	42	45	18	29	134	7	91	36	5	33	165	116	77	55	11	43	186
20	45	50	20	50	165	8	60	0	0	0	60	40	54	40	26	36	156
19 229	58	47	5	42	152	8	100	46	0	8	154	57 741	82	47	13	51	193
	51	61	9	37	158	28	87	29	7	15	138		71	55	10	44	180
189	50	57	8	33	148	35	88	28	9	16	141	646	71	55	7	46	179

S. borealis in grass samples in the spring 1977 (Table 4). In this year growth of the fungus was observed under the snow on leys at the Lapland experimental station as early

as the end of March. At the end of April there was abundant growth of the fungus and also destruction of the grass. However, large amounts of water from melting snow

Table 4. Frequency of Sclerotinia borealis, number of sclerotia per sample, and size of sclerotia in the grass sample.

		Scleroti	a no. per	sample		Distr	ribution o	of sclero		ding by	size,
Localities	1976	1977	1978	1979	1976—		1978			1979	
	1310	20			1979	$< 1 \atop  ext{mm}$	1—3 mm	> 3 mm	$< 1 \  m mm$	1—3 mm	> 3 mm
								10	c	61	33
Tornio	27	27	5	46	26	33	50	17	6		24
Ylitornio	59	101	152	86	100	34	48	18	16	60	100000000000000000000000000000000000000
Tervola	4	86	66	49	51	40	42	18	1	70	29
Rovaniemi	27	68	19	48	41	25	65	10	7	72	21
Kemijärvi		14	23	51	29	39	48	13	22	64	14
Salla	45	32	7	51	34	42	56	2	25	61	14
Pelkosenniemi	_	157	5	28	63	40	59	1	43	43	14
Sodankylä	43	146	17	51	64	50	44	6	4	76	20
•	36	98	40	56	58	55	35	10	4	78	18
Kittilä	30	45	13	38	32	64	29	7	26	50	24
Muonio Inari	33	53	144	70	75	74	24	2	29	60	11
Mean	34	75	45	52	52	22	18	5	8	33	10
Portion <sup>0</sup> / <sub>0</sub>						45	45	10	17	63	20

in the beginning of May stopped the fungal growth on many leys before the final disappearance of snow on 15.5. In other places the fungus continued to grow under thick, slowly-melting snow drifts. Also at Muddusniemi extensive growth of *S. borealis* was observed at the end of March and beginning of April in samples taken from under the snow in the spring of 1961 (Nissinen and Salonen 1972 a). It is obvious that the longer the fungus has managed to grow in the grasses, the greater is the injury (cf. Pohjakallio and Salonen 1956).

One important characteristics of *S. borealis* was that it occurred abundantly in some leys and scantily in others. It was found most commonly and abundantly in young timothy leys, especially first-year leys (cf. Pohjakallio and Salonen 1956, Nissinen and Salonen 1972 a, Blomqvist 1970, Valmari 1979). It appeared in leys established both in spring and in autumn. In older stands which were thick and profuse the fungus could cause small sharply-defined round patches. These patches could also be of different sizes and shapes, or, on the other hand, the fungus could sometimes occur uniformly throughout the whole

ley (Plate I A). The field could also be completely destroyed. In young stands sown in the late summer or autumn, the fungus killed the tender plants usually along certain rows or in variously-sized patches. In such young stands the sclerotia were considerably smaller than in the older and more dense stands.

Sclerotia of S. borealis could sometimes occur in the spring in very great numbers, and could be literally gathered by handfuls. This was especially true when in the previous autumn a vigorous stand was left uncut beneath the snow. Nevertheless, the ley in the following summer could have a good growth. In such cases the damage had occurred only to the leaves, while the roots remained healthy. In other cases where the fungus had also destroyed the roots, there could be very great damage (cf. Pohjakallio and Salonen 1956). Sparse stands of grass could also be destroyed by the fungus, although sclerotia did not develop as abundantly as in vigorous stands.

Sclerotia of S. borealis occurred in the leaves, the sheaths and the root collars of the plants, depending on the species of grass (Fig. 9, Plate IB). They could also be found

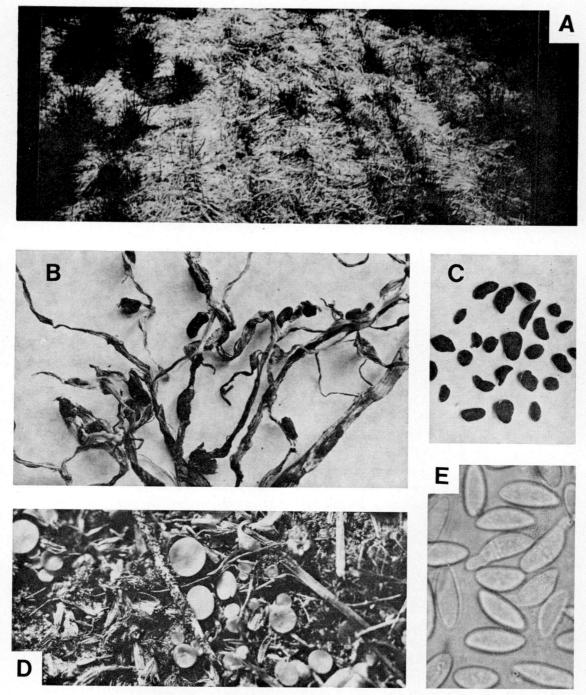


Plate I. Sclerotinia borealis on Phleum pratense. A: A damaged ley in the spring. B, C: Sclerotia of the fungus, B: on the dead leaves. D: Apothecia of the fungus on soil surface of the first-year timothy ley at Apukka in 13. 10. 1980. E: Ascospores of the fungus. B, C, D: X 1. E: X 1000.



Fig. 9. The dead first-year grasses damaged by *Sclerotinia borealis*. A: Festuca pratensis, B: Poa annua, C: Poa pratensis, D: Poa alpigena, E: Festuca rubra, F: Dactylis glomerata. Matherial: A, D, E: InL. Inari, Muddusniemi 13. 6. 1979. B, F: KemL: Kittilä 27. 5. 1976. C: PP: Rovaniemi, Apukka 15. 6. 1979. A-F: X 1.

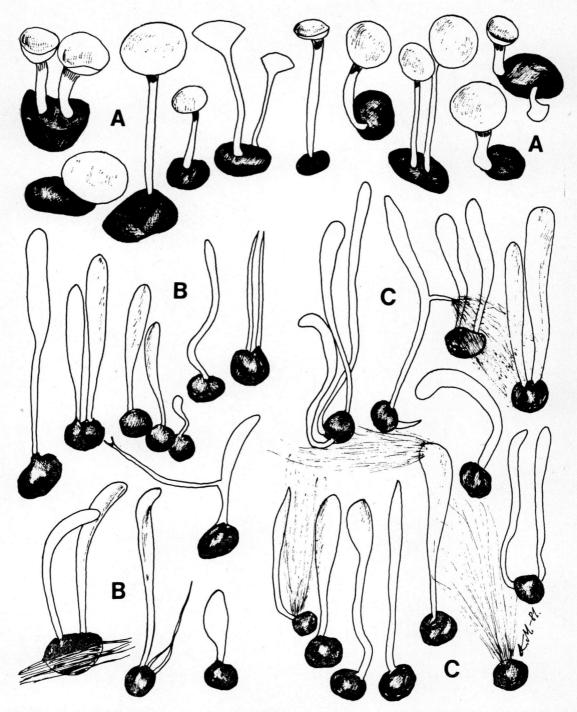


Fig. 10. A: Apothecia developed in sclerotia of *Sclerotinia borealis* on first-year timothy ley at Rovaniemi, Autti 15. 10. 1980. B, C: Sporophores of *Typhula ishikariensis* on first-year timothy ley, B: Sodankylä, Vaalajärvi 14. 10. 1980, C: Rovaniemi, Apukka 13. 10. 1980. A: X 4. B, C: X 5.

Table 5. Occurrence of sclerotia of *Sclerotinia borealis* at different depths in the soil, spring 1978 (ley sample  $10 \times 10 \times 10$  cm).

Localities	No. of the leys	No. of the samples	S. borealis no. from samples	depth in	t different the soil, ean
Document	investigated	investigated	no. Irom samples	0—1,5 cm	1,5—5 cm
	4	6	0	0	0
Tervola	10	15	q	112,7	0,3
Rovaniemi	10	15	9	5,6	0,6
Salla	4	9	9	10,5	0,0
Pelkosenniemi	2	2	2		0.3
Sodankylä	4	4	2	51,8	0,3
Kittilä	2	3	3	151,3	0
Muonio	1	1	1	157,0	0
Inari	i	ī	1	95,0	0
Total	28	37	21		
Mean				73,0	0,1

lying on the ground where they had fallen from the plants. In young first-year growths the sclerotia were easy to see. In contrast, older leys often had abundant dead leaves, stubble and other plant debris. This made it difficult to observe the sclerotia both in the grass and on the ground (cf. Nissinen and Salonen 1972 a).

The present study included newly-cleared land, both on mineral and on peat soils. Even in these completely new leys damage caused by *S. borealis* could be seen already after the first year. The same observation was made at the North-Savo experiment station in one field of winter rye on virgin peat soil in 1947 (Jamalainen 1949), as well as in the 1950s on newly cultivated land at the Muddusniemi experimental farm (Pohjakallio and Salonen 1956).

# Location of Sclerotinia borealis sclerotia in the soil

The location of sclerotia of S. borealis at various depths in the soil was studied in ley samples taken from both peat and mineral soils in different areas (Tables 5 and 6). Almost all the sclerotia, averaging  $98\,^{0}/_{0}$  (geographical variation 90— $100\,^{0}/_{0}$ ), were found

on the surface of the ground or in the uppermost 1,5 cm of the soil. Only a very few sclerotia were encountered down to a depth of 5 cm. This situation was similar for all the leys investigated.

Preservation of *Sclerotinia borealis* sclerotia in the soil and development of apothecia in them

In the years 1978—1980 studies were made in various locations (Inari, Kittilä, Rovaniemi, Maaninka, Tikkurila) on the preservation of sclerotia of *S. borealis* and the development of apothecia, both on the ground surface and at a depth of 5 cm (Table 7). The sclerotia remained unchanged during the winter season 17.—20. 10. 1978—13.—15. 6. 1979. Of the sclerotia on the soil surface 0—5% were destroyed during the winter, while 13—20% of those at a depth of 5 cm were destroyed.

One year later (16.—29.10.1979) the sclerotia on the soil surface were still in good condition. Those on the surface of mineral soil had  $0-15\,\%$  losses and on peat soil 5—14 % losses. In contrast, many more of the sclerotia at a depth of 5 cm were destroyed,

Table 6. Occurrence and size of sclerotia of *Sclerotinia borealis* in the soil, spring 1979 (ley sample  $10 \times 10 \times 10$  cm).

Localities	No. of the leys investigated	No. of sclerotia per sample		Distribution of sclerotia according by size, % per sample			
	mvestigated	mean	range	0—1 mm	1—3 mm %	3 mm %	
Tornio	1	28	28	3,5	85,8	10,7	
Tervola	4	6	0—8	0	66,7	33,3	
Rovaniemi	2	10	7—12	40,0	50,0	10,0	
Kemijärvi	2	7	0-14	14,3	85,7	0	
Sodankylä	4	0			_	_	
Kittilä	2	37	36-38	5,4	62,2	32,4	
Muonio	7	6	0-26	16,7	66,7	16,7	
Total	22	2 2	27				
Mean		13,4	0-38	10,5	66,9	22,6	

averaging 30  $^{0}/_{0}$  in mineral soil (variation in the different areas 3—52  $^{0}/_{0}$ ) and 15  $^{0}/_{0}$  in peat soil (14—23  $^{0}/_{0}$ ).

Two years later (14.—29.10.1980) there were still well-preserved sclerotia on the ground surface. On mineral soils many more had been destroyed (av.  $52\,^{0}/_{0}$ , range 34— $66\,^{0}/_{0}$ ) than on peat soils (av.  $13\,^{0}/_{0}$ , range 0— $17\,^{0}/_{0}$ ). At a depth of 5 cm the sclerotia in peat soil had disappeared completely and in mineral soils the losses averaged  $74\,^{0}/_{0}$  (range 24— $100\,^{0}/_{0}$ ). Only a few disintegrated remains were to be seen. In the studies of Årsvoll (1976) dry sclerotia remained germinationable viable at room temperature for over three years.

In both of the autumns 1979 and 1980 apothecia developed only in the sclerotia which were on the ground surface. In the autumn 1979 apothecia developed in an average of  $17^{\circ}/_{\circ}$  of the sclerotia (variation among the different areas  $0-68^{\circ}/_{\circ}$ ). In the autumn 1980 fewer apothecia developed, averaging  $8^{\circ}/_{\circ}$  (range  $0-43^{\circ}/_{\circ}$ ).

Muddusniemi was the only trial location where apothecia did not develop. In contrast, as far south as Tikkurila apothecia developed in both autumns in about 10 % of the sclerotia. These were small in size, averaging only

1,5 mm in diameter. Also in the laboratory studies of Årsvoll (1976) sclerotia on the soil surface had a germination of as high as  $93.8~^{0}/_{0}$ , while at a depth of 10 mm only 16,5 germinated, and at 20 mm depth there was no germination at all.

The occurrence of apothecia of S. borealis under field conditions was investigated in the month of October 1980 (13.-16.10.) at the Lapland experimental station and at 10 farms in the communes of Rovaniemi, Sodankylä and Tervola (Table 8, Plate ID, Fig. 10 A). In this material, which comprised a total of 125 trial plots on 30 leys, there were an average of 24 apothecia per 225 cm2 (range on the different leys 1—200). The variation was very great between the different leys. According to observations made at Muddusniemi (Pohjakallio and Salonen 1956) in the autumn 1955 apothecia began to develop around the 10th of September. After this date they occurred very abundantly in the leys. In a meadow fescue stand which was sown in 1951, an average of 43 apothecia per m<sup>2</sup> were observed on 5.10.

In the present study leys which were established in 1980 were generally found not to have any apothecia or at the most they were found only in extremely small amounts.

Table 7. Preservation of sclerotia of *Sclerotinia borealis* on soil surface and at a depth of 5 cm in the different localities, 1978—1980. Sclerotia were in wire netting bag, 100 per bag.

Localities	Soil	Depth <sup>1</sup>	Date of beginning	Date of terminating	Total Apothecia developed Ruined	Strains of the fungus
Inari, Muddusniemi 69°04'N, 27°03'E	Mineral soil	Surface 5 cm	20. 10. 78	13. 6. 79 — » —	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	Muddusniemi 197
	» »	Surface 5 cm	20. 10. 78 — » —	29. 10. 79 — » —	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	»
	» »	Surface 5 cm	20. 10. 78	28. 10. 80 — » —	34 0 66 17 0 83	_ » _ » _ »
Kittilä 67°41'N, 24°54'E	Mineral soil	Surface 5 cm	20. 10. 78 — » —	23. 10. 79 — » —	$\begin{array}{cccc} 100 & 10 & & 0 \\ 48 & 0 & 52 \end{array}$	Muddusniemi 197
	Peat soil	Surface 5 cm	_ » _	_ » _ _ » _	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	— » — Kittilä 1978
	Mineral soil	Surface 5 cm	20. 10. 78 — » —	24. 10. 78 — » —	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	»
	Peat soil	Surface 5 cm	20. 10. 78 — » —	24. 10. 80 — » —	$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	— » —
Rovaniemi, Apukka 66°35'N, 26°00'E	Mineral soil	Surface 5 cm	17. 10. 78	15. 6.79	$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	Muddusniemi 197
	Peat soil	Surface 5 cm	»	»	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	
	Mineral soil	Surface 5 cm	17. 10. 78	20. 10. 79	86 5 14 56 0 44	»
	Peat soil	Surface 5 cm	»	» »	86 26 14 86 0 14	
	Mineral soil	Surface 5 cm	17. 10. 78	20. 10. 80	60 0 40 36 0 64	» »
	Peat soil	Surface 5 cm	17. 10. 78	20. 10. 80 — » —	100 43 0	» »
Maaninka, Halola 63°09'N, 27°19'E	Mineral soil	Surface 5 cm	17. 10. 78 — » —	16. 10. 79 — » —	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	Tervola 1978
	»	Surface 5 cm	17. 10. 78 — » —	14. 10. 80 — » —	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	
Tikkurila 60°17'N, 25°04'E	Mineral soil	Surface 5 cm Surface	17. 10. 78 — » — 17. 10. 78	19. 10. 79 — » — 29. 10. 80	85 10 15 77 0 23 66 11 34	Muddusniemi 197
	»	5 cm	— » —	»	0 0 100	_ » _

Surface = On soil surface 5 cm = At a depth of 5 cm

Similarly only a few apothecia occurred in older third- and fourth-year leys. The largest numbers of apothecia were found in first-and second-year stands.

In addition to germinated sclerotia, there were also ungerminated sclerotia on the ground surface. Often these two categories occurred approximately in the same proportions, either few of both kinds or many of both kinds.

The average diameter of a total of 525 apothecia of *S. borealis* was found to be 3,2 mm (range 1—8 mm). This figure was similar on most of the leys. According to Pohjakallio and Salonen (1956) the diameter of apothecia found in leys was 1—6 mm. The studies of Årsvoll (1975) in Norway gave a corresponding figure of 1,8 (2—6) mm.

Examinations of 460 sclerotia found on different leys showed that in general only one

Table 8. Occurrence of apothecia of Sclerotinia boerealis in some leys in North Finland, 13.—16. 10. 1980.

Localities	Farm	Ley Soil	Soil	Age of ley year	No. of a	No. of apothecia per 225 cm <sup>2</sup>	
					mean	range	established leys +
Sodankylä	1	1	mineral	2	6,8	4—16	+
Vaalajärvi		2	»	01	1,3	1—2	<u>.                                     </u>
		3	— » —	3	9,2	1—27	+
		4	peat	4	8,3	3—20	÷
	2	5	mineral	4	9,1	4-20	+
		6	— » —	2	11,3	2-40	+
		7	— » —	1	11,3	2-39	+
	3	8	— » —	2	18,5	11—30	<u> </u>
Rovaniemi	4	9	peat	1	130,0	96—155	+
Apukka		10	— » —	1	66,0	66	+
		11	mineral	2	27,0	10—60	+
	5	12	»	4	23,0	8—38	<u> </u>
		13	— » —	01	1,0	1	
		14	— » —	1	11,5	2—37	<u> </u>
Rovaniemi	6	15	— » —	4	8,5	2-20	<u>.                                     </u>
Autti		16	— » —	3	9,0	2—19	<u> </u>
		17	— » —	2	16,2	6—26	<u> </u>
		18	»	1	92,2	23-200	
		19	»	1	43,6	10—84	<u></u>
		20	peat	1	30,4	16—45	<u></u>
		21	- » —	2	41,7	30—58	
	7	22	mineral	2	13,3	2—52	_
Tervola	8	23	peat	1	2,3	1—3	
		24	mineral	2	4,0	2—6	_
	9	25	— » —	1	20,0	20	
		26	— » —	2	23,0	23	:
	10	27	peat	1	10,0	10	
		28	mineral	1	27,0	10—35	_
	11	29	— » —	2	27,3	10—42	
	12	30	— » —	2	2,0	2	-
Total	12	30		1000			1
Mean					23.5	1-200	

<sup>1</sup> established in the summer 1980

apothecium developed from one sclerotium. This occurred in  $95\,^{0}/_{0}$  of the cases (range  $80\,$ — $100\,^{0}/_{0}$ ). Two apothecia in the same sclerotium occurred in  $4\,^{0}/_{0}$  of the cases, while three and four apothecia were found only in rare instances. In the material of Årsvoll (1975) 1—6 (1—3) apothecia developed from one sclerotium.

Speckled snow mould (Typhula spp.)

In Finland Typhula incarnata and T. ishikariensis occur commonly throughout the whole country in winter cereals and in many kinds of ley grasses (Jamalainen 1957).

There are only scanty data concerning the occurrence of *Typhula* species in leys in North Finland, as well as the relative proportions of these species. However, *Typhula* spp. has been mentioned together with *Sclerotinia borealis* and *Fusarium nivale* as being among the most pathogenic low-temperature fungi in these regions (Pohjakallio and Salonen 1956, Jamalainen 1970, 1978). At Muddusniemi *Typhula* spp. was especially prevalent in ley grasses in the spring 1950, when the snow melted earlier than usual and

<sup>&</sup>lt;sup>2</sup> pentachloronitrobenzene, PCNB

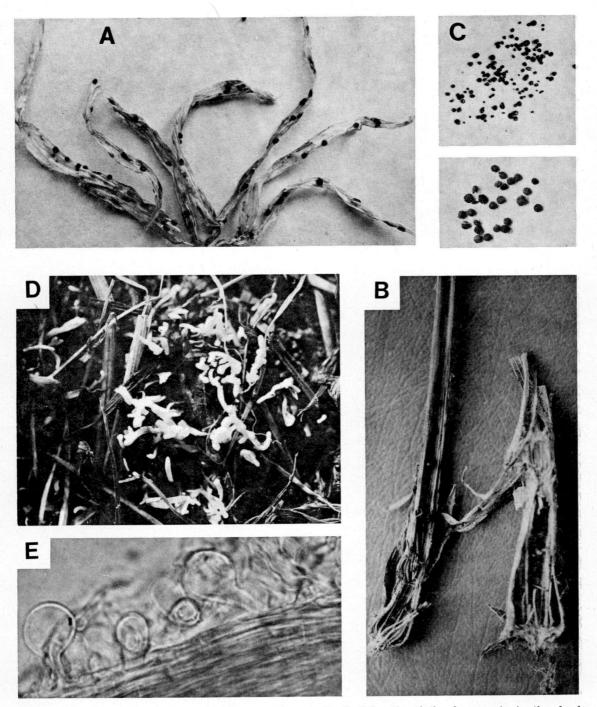


Plate II. Typhula ishikariensis on Phleum pratense. A, B, C: Sclerotia of the fungus, A: in the dead leaves, B: in the dead culms and the enlarged basal part of the stem. D: Sporophores of the fungus on soil surface of the timothy ley at Tervola in 16.10.1980. E: Basidia of the fungus. A, B, C, D: X 1. E: X 1000.

spring frosts damaged the plants. At the Lapland experimental station *T. ishikariensis* has caused injuries in many winters to different grass species (Jamalainen 1970, 1978). Older leys appeared particularly to suffer from such injuries (Valmari, oral commun.).

In the present investigation T. ishikariensis was the most common of the Typhula species. In the entire material it occurred in  $55\,^{0}/_{0}$  of the samples (variation among the communes  $38-75\,^{0}/_{0}$ ). The fungus was more common in the southern and central parts of the region than in the northern parts.

In the years 1976, 1977 and 1978 the fungus was rather prevalent, occurring in an average of 61—69% of the leys. In contrast, in the spring 1979 the fungus was found in only 29% of the leys. A partial reason for this may be the spring rains, since the sclerotia of *T. ishikariensis* when growing on the surface of the plant are very easily washed off.

T. ishikariensis occurred particularly in older timothy leys in which it could cause extensive damage. The fungus produced different-sized brown patches in stand. Sclerotia developed generally in the leaves (Plate II A). If the fungus injured only the leaves, the damage was minor. Sclerotia could also be in the culms and the enlarged basal part of the stem, and in these cases the plants had been killed (Plate II B). Such damage occurred especially in the springs of 1977 and 1979, when many leys were destroyed, particularly at Sodankylä and Kittilä.

Germination of *T. ishikariensis* in the field was observed abundantly in the autumn 1980. In this year the weather remained warm longer than usual, until the middle of October. Pale, skin-coloured sporophores grew generally in dense clusters (Plate II D, Fig. 10). This was due to the fact that the sclerotia most often occurred in rows in the tissues of the leaves and stems. In the same sclero-

tium usually one sporophore arose, often two, sometimes even three (cf. Årsvoll 1975). The average length of 120 sporophores examined was 4,7 mm (range among different leys 3,8—6,0 mm and among individual sporophores 2—12 mm). The thickness of the sporophores varied from 0,5 to 1,0 mm. In the studies of Årsvoll (1975) the sporophore length was 5—20 mm and the thickness 0,5—1,0 mm. Under moist conditions abundant growth of white mycelia readily took place. Also basidiospores germinated readily.

Typhula incarnata was found in the whole material in an average amount of only  $10\,^{\circ}/_{\circ}$  (variation between the years  $3-15\,^{\circ}/_{\circ}$ ). The fungus was most common in leys in the spring 1977. Compared with T. ishikariensis, the significance of T. incarnata was minor.

In Norway both of these *Typhula* species are prevalent in most of the country. *T. ishikariensis* is the most important winter-damage fungus in areas where the snow cover persists longer than 150 days and where there are at least 80 days with an air temperature under 0°C. *T. incarnata* is most common in areas with a mild and moist winter climate and where the snow cover lasts for at least 90 days. The injuries caused by this species are generally light. Even severely-infected plants often recover (Årsvoll 1973, 1975).

In Iceland *T. incarnata* occurs widely in the northern parts of the country, but it generally causes only minor injuries. On the other hand, *T. ishikariensis* apparently causes extensive damage, even though it is less prevalent than *T. incarnata* (Kristensson and Gudleifsson 1976).

#### Snow mould (Fusarium spp.)

Snow moulds occurred quite commonly throughout the entire region of this study, amounting to an average of 44% in the

There were considerable whole material. variations between the years. In the spring of 1976 and 1977 Fusarium species were found in an average of 63 % and 66 % of the leys examined, whereas in 1978 and 1979 they were much less prevalent, averaging  $37 \, ^{\text{0}}/_{\text{0}}$  and  $15 \, ^{\text{0}}/_{\text{0}}$  of the samples. In general, they occurred only in small amounts in the leys. In timothy they were not particularly significant as a cause of winter injury. On the other hand, meadow fescue appeared to be much more susceptible to damage by Fusarium species. Especially susceptible was English ryegrass which could be totally destroyed by these fungi (cf. Ylimäki 1955). Likewise at the Lapland experimental station (Jamalainen 1970) and in Norway (Arsvoll 1973) F. nivale produced greater damage in Lolium perenne than in the other grasses. There were several species of Fusarium (Table 8). The most common were Fusarium avenaceum (av. 11 %), F. semitectum (av.  $8^{0/0}$ ), F. nivale (av.  $7^{0/0}$ ), and F. culmorum (av. 4 %). In addition, sporadic occurrences were also observed of F. graminearum, F. moniliforme, F. oxysporum, F. poae and F. tricinctum.

Winter injuries caused by snow moulds in levs in Lapland have occurred only rarely and then only in small amounts, even though damage to winter cereals can be quite great. For example, during the trial period 1947-1955 at the Muddusniemi experiment farm Fusarium species occurred to some extent in ley grasses only in the years 1950 and 1951, whereas in the other years they scarcely appeared at all (Pohjakallio and Salonen 1956). Likewise at the Lapland experiment station during the period 1951-1977 F. nivale was found abundantly in timothy, cocksfoot and meadow fescue only in the spring 1967, and even then it did not cause appreciable damage (Jamalainen 1970, 1978).

On the other hand, in Norway F. nivale

Table 9. Frequency of the most important parasitic fungi, mean  $^{0}/_{0}$  of the samples and variation by localities.

localities.				
	Frequency, no%			
Fungi	of the samples	variation by localities		
As comy cotina				
Pyrenomycetes  Womographella minglis	0.1	0 1		
monographena mouns	0,1	0— 1 .		
(Rehm) E. Müller				
Loculoascomycetes		0—10		
Phaeosphaeria herpotri-	3	0—10		
choides (de Not.) L. Holm				
Discomycetes	17.1	51—84		
Sclerotinia borealis	71	21—04		
Bubàk & Vleugel				
Basidiomycotina				
Holobasidiomycetidae	19	10—42		
Rhizoctonia solani Kühn	19	10 12		
Amphyllophorales	10	0—26		
Typhula incarnata Lasch ex Fr.	10	0 40		
T. ishikariensis Imai	55	38—75		
	00	00 10		
Deuteromycotina \				
Hyphomycetes Alternaria tenuis auct.	2	0-14		
Botrytis cinerea Pers. ex Fr		0_29		
<del>-</del>	. 13 70	0—29 44—83		
Cladosporium spp.	5 .	113		
Epicoccum purpurascens	υ.	1 10		
Ehrenb. ex Schlecht	27	1140		
Fusarium spp.	11	11—40 7—17		
F. avenaceum (Corda	11	1		
ex Fr.) Sacc. F. culmorum (W. G.	4	8 —0		
Smith) Sacc.	*	0 0		
F. graminearum	1	0— 4		
Schwabe	•	0 1		
F. nivale (Fr.) Ces.	7	0-13		
F. semitectum Berk. &	8	0 <b>—13</b> 0—19		
Rav.	Ū	0 20		
Helminthosporium spp.	6	015		
Heterosporium phlei	5	0-15		
Gregory	U	0 10		
Pestalotia truncata Lev.	0,3	0— 2		
Pseudocercosporella	1	$0 - \frac{1}{2}$		
herpotrichoides (Fron)	-	· -		
Deighton				
Trichoderma viride	33	1653		
Pers. ex S.F. Gray	Ų.	20 00		
· Volucrispora graminea	0,3	0 3		
Ingold, McDougall &	0,5	, ,		
Dann				
Coelomycetes				
~ 1 1 1				
Ascochyta spp.	3	0—10		
Coniothyrium sp.	6	1—17		
Hendersonia crastophila	2	0—10 1—17 0— 6		
Sacc.	. 4	<b>~</b> 0		
H. culmicola Sacc.	1	0—10		
Phaeoseptoria festucae	$\overset{1}{2}$	0—22		
	4	0-22		
Sprag.	12	0—49		
Phoma spp.	0,4	0-49		
Mycelia sterilia	0,4	<u> </u>		
(Basidiomycete)				
Bacteria				
Actinomycetales	41	14—58		
Streptomycetes spp.	-11			
		12		

is the most important low-temperature parasitic fungus in all grass species and is found throughout the whole country (Årsvoll 1973, 1975). Among other Fusarium species F. avenaceum also occurs prevalently as a pathogen on grasses, while F. culmorum and F. semitectum are less significant (Årsvoll 1975).

# Other fungi occurring in leys

In addition to Sclerotinia borealis, Typhula spp. and Fusarium spp., a large number of other less pathogenic fungi were found in

the grass samples (Table 9). The most common of these were Cladosporium spp., Trichoderma viride, Rhizoctonia solani, Botrytis cinerea and Phoma spp.

Most of these same fungi are found in leys in Norway. In laboratory experiments these fungi have been demonstrated to be pathogenic, at least to some extent (Årsvoll 1975). The bulk of these cosmopolitan species are considered to be generally saprophytic or weakly parasitic (Domsch and Gams 1970, Ellis 1971). The total numbers of fungi found in grass samples in the present study were 75 species belonging to 64 genera.

#### CONCLUSIONS

The original incentive for starting the present study was the period of extensive damage occurring in the 1970s to leys in North Finland. Farms with the most damage to leys were initially chosen for the investigation. In later years attempts were made to obtain a more generalized picture of the leys by including all kinds of farms. Despite these attempts, however, the material apparently gives a less favourable view of northern Finnish leys than the average. The material has perhaps a lack of uniformity which was due to the short life of the leys, the variations in quality of the farms and the farmers, etc. The material is somewhat scanty, and the time spent on the investigation as well as the other possibilities did not correspond to the extent of the subject and the geographical area.

The advantage of this kind of material is its diversity. In cases where the intention is to make a general overall survey of the situation in the whole region, e.g. by investigating the fungal population, this method has been generally used (cf. Jamalainen 1970, Årsvoll 1973). In Norway winter damage

to leys was comprehensively studied in the 1970s (e.g. Årsvoll 1973, 1975, 1976, 1977, Årsvoll and Larsen 1977). A large part of these excellent studies can be applied also to conditions in Finland.

The snow cover in North Finland is thick and it often lasts from October to May (Kolkki 1966). Under the snow cover there is darkness, sufficient moisture and a temperature remaining around 0°C (Ylimäki 1962). Many of the low-temperature parasitic fungi grow at temperatures between 0°C and -6°C, and in other respects, too, they drive under such conditions (Ekstrand 1955, Pohjakallio and Salonen 1956, Jamalainen 1970, Nissinen and Salonen 1972 a, Årsvoll 1975). On northern Finnish leys these fungi therefore have a suitable environment for growing throughout the whole winter. However, most of the damage caused by these fungi occurs in the latter part of the winter and the spring (cf. Pohjakallio and Salonen 1956. Nissinen and Salonen 1972 a). Also in the present study it was found in ley samples taken from beneath the snow that S. borealis and Typhula ishikariensis produced mycelia

and young sclerotia no earlier than the latter half of March, and this occurred only in 1977. In this year the fungi caused most of the damage under thick, slowly melting snow drifts (cf. Pohjakallio and Salonen 1956).

The great influence of weather conditions on the occurrence of winter damage and the annual variations are generally known (Ekstrand 1955, Pohjakallio and Salonen 1956, 1958, Jamalainen 1970, 1978, Nissinen and Salonen 1972 a, Årsvoll 1973). Also in this study weather factors were of profound significance. The greatest amounts of poorly-overwintered leys occurred in the spring 1977 after a long winter with a deep snow cover. The least winter damage was found in the spring 1979 after a winter with a thinner snow cover and lower temperatures than usual (cf. p. 112).

The total amounts of the most serious low-temperature pathogenic fungi Sclerotinia borealis, Typhula ishikariensis and Fusarium spp. were found to vary in the samples investigated in a way corresponding to the extent of injury to the ley.

These observations confirm the fact that in the present study low-temperature parasitic fungi were the major cause of winter damage to leys in North Finland (cf. Ekstrand 1947, 1955, Jamalainen 1970, 1980, Ylimäki 1955, Pohjakallio and Salonen 1956, Nissinen and Salonen 1972 a, Årsvoll 1973, 1975).

In most cases the amount of winter damage to first-year timothy leys has been directly proportional to the incidence of *S. borealis* (cf. Isotalo and Vogel 1962, Nissinen and Salonen 1972 a).

The significance of age of the ley as regards the occurrence of low-temperature parasitic fungi is generally known (Pohjakallio and Salonen 1956, Jamalainen 1970, Årsvoll 1973, 1977, Valmari 1979). Also in this study the injuries were greatest in first-year timothy leys. This is due partly to the slow initial growth

of the young timothy plants, especially their root growth (Salonen 1951, Jäntti 1953). These fungi, in particular S. borealis, first destroy the foliage and then continue their parasitic growth in the roots. In older timothy stands it is more difficult for the fungus to destroy the roots than in young stands (Pohjakallio and Salonen 1956). vigorous stand of timothy in the autumn of its first season offers a better environment for the growth of parasitic winter fungi than does an older ley with its often dry, withered stubble (Nissinen and Salonen 1972 a). According to the view held by many farmers, the stubble of a companion crop for the same reason reduces fungal damage in many leys.

Many studies have shown that grass varieties originating in northern areas are more resistant to winter fungi than varieties of southern origin (Ekstrand 1955, Pohjakallio and Salonen 1956, Andersen 1971, Jamalainen 1970, 1974, Nissinen and Salonen 1972 a, Årsvoll 1977). The situation today, however, is that in northern Finland grass varieties intended for use in southern areas are being grown and even their seed is being produced.

According to many investigators (Huokuna 1971, Huokuna and Hiivola 1974, Pulli 1980) too heavy nitrogen fertilization weakens the winter resistance of leys. At the same time injuries caused by low-temperature parasitic fungi increase (Ekstrand 1955, Nissinen and Salonen 1972 b). In laboratory studies by Arsvoll and Larsen (1977) increasing amounts of nitrogen fertilization caused a reduction in resistance of grasses to Fusarium nivale, Typhula ishikariensis and Sclerotinia borealis, and concurrently their frost durability was also reduced. By decreasing the nitrogen levels on leys, the Lapland experimental station has in recent years achieved a large increase in winter dependability (Valmari Similar results have also been obtained by many farmers. On the other hand, nitrogen fertilization has improved the resistance of timothy to *S. borealis* (Jamalainen 1970), namely in the latest sowings (Nissinen and Salonen 1972 b).

Increasing the amount of phosphorus has generally improved the winter durability of leys, especially against the most important winter fungi (Ekstrand 1947, Årsvoll and Larsen 1977).

Hardening of plants is very important in regard to their winter durability (Årsvoll 1975, 1977, Tronsmo 1980). Associated with

this hardening process are chemical changes in the plant tissues (Pohjakallio et al. 1959, Arsvoll and Larsen 1977, Pulli 1980). The amount of food reserves in the plant roots, above all the sugar content, is a measure of its winter durability (Pohjakallio et al. 1959, Huokuna 1971, Vestman 1980). Two factors affecting this are the fertilization and the last cutting of the ley (Juola et al. 1977, Hakkola 1978, 1980, Huokuna 1980). Much research work remains to be done on these problems concerning the leys in North Finland.

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#### SELOSTUS

# Pohjois-Suomen nurmien talvituhoista ja talvituhosienistä vuosina 1976-1979

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#### Maatalouden tutkimuskeskus

Aineisto käsitti 740 peltoa 11 pitäjän alueelta. Näytteet kerättiin keväisin tehdyillä tarkastusmatkoilla. Tutkimuksia täydennettiin viljelijäin haastatteluilla. Sienet tutkittiin laboratoriossa mikroskooppisesti.

Tutkituista nurmista oli valtaosa, <sup>3</sup>/<sub>4</sub>, timoteinurmia, timotei-nurminata ja timotei-pohjannurmikka -sekanurmia oli kumpiakin 10 <sup>9</sup>/<sub>0</sub>. Nurmista sijaitsi <sup>3</sup>/<sub>4</sub> kivennäismailla ja loput turvemailla. Iältään olivat nurmet valtaosin nuoria, 1. ja 2. vuoden nurmia. Noin puolet nurmista oli perustettu ilman suojakasvia. Suojakasvina olivat yleisimpiä ohra ja kaura. Esikasvina oli miltei aina peltonurmi.

Huonosti talvehtineita nurmia oli eniten keväällä 1977 ja vähiten keväällä 1979. Yleisimmät talvituhosienet olivat pohjanpahkasieni (Sclerotinia borealis), pahkulasienet (Typhula spp.) ja lumihomeet (Fusarium spp.) Nämä sienet yhdessä olivat yeisempiä Lapin itä- ja pohjoisosissa kuin länsi- ja eteläosissa. Vuosittainen vaihtelu oli myös

hyvin samanlainen nurmien talvituhojen esiintymisen kanssa. Keväällä 1977 näitä sieniä oli eniten, keväällä 1979 vähiten.

- *Ṣ. borealis*ta esiintyi tässä aineistossa keskim. 71 % nurmista. Se oli yleisempää pohjoisessa (Sodankylä, Kittilä, Inari) kuin eteläisimmillä alueilla.
- S. borealista esiintyi yleisimpänä ja runsaimpana 1. vuoden timoteinurmissa, joissa sieni aiheutti suurimmat tuhot. Myös vastaraivatussa uudismaassa saattoi tuhoa tavata jo 1. vuonna. Eri aikaan ja eri tavoin perustettujen nurmien, sen paremmin kuin kivennäis- tai turvemaalla kasvavien nurmienkaan välillä ei juuri johdonmukaisia eroavuuksia havaittu. Sen sijaan viereisissä, yksittäisissä nurmissa saattoivat tuhot olla hyvinkin erilaisia. Suuria olivat nimenomaan vuosien väliset erot.

Keväällä 1977, jolloin S. borealisen tuhot olivat suurempia kuin muina tutkimusvuosina, sieni kasvoi nurmissa lumen alla jo maaliskuun loppupuolelta lähtien. Sen sijaan keväällä 1979, jolloin talvituhoja oli vähiten, ei myöskään sienen kasvua lumen alla todettu.

S. borealisen pahkat sijaitsivat nurmissa paitsi kasvustossa miltei yksinomaan maan pinnalla tai aivan pintakerroksessa. Pahkat säilyivät luonnossa, nurmessa maan pinnalla elävinä ja itämiskykyisinä ainakin kaksi vuotta. Sen sijaan niistä 5 cm syvyydessä tuhoutui kivennäismaalla 3/4 ja turvemaalla kaikki. Myös kotelomaljoja kehittyi vain maan pinnalla olleisiin S. borealisen pahkoihin.

Mustapahkulasientä (Typhula ishikariensis) esiintyi koko aineistossa, keskim. 55 %. Sieni oli

yleisempi Lapin eteläisissä ja keskisissä osissa kuin pohjoisemmilla nurmilla. Sieni aiheutti tuhoa varsinkin vanhemmissa timoteinurmissa. Ruskopahkulasienen (*Typhula incarnata*) merkitys jäi vähäiseksi.

Lumihomeita (Fusarium-lajeja) esiintyi kautta alueen, keskim. 44 % tutkituista nurmista. Niitten määrä oli nurmissa yleensä pieni. Varsinkin timoteilla sienten merkitys jäi vähäiseksi. Nurminata näytti olevan alttiimpi. Fusarium-lajeista olivat F. nivalen ohella yleisimpiä F. avenaceum, F. culmorum ja F. semitectum.

Lisäksi nurmissa todettiin suuri määrä muita sieniä, yhteensä noin 75 lajia, kuuluen 64 sukuun.

# Seria PHYTOPATHOLOGIA N. 79 — Sarja KASVITAUDIT n:o 79

# ON THE SEED-BORNE MICROFUNGI ON WILD GRASSES IN FINLAND

#### KAIHO MÄKELÄ

Mäkelä, K. On the seed-borne microfungi on wild grasses in Finland. Ann. Agric. Fenn. 20: 132—155 (Agric. Res. Centre, Inst. Pl. Path. SF-01300 Vantaa 30, Finland).

The fungal microflora of 63 species of wild grass seed were determined from 284 samples collected throughout Finland, but mostly from southern parts of the country. Over 90 species of fungi, belonging to 60 genera, were observed. The fungi included three species of Pleospora including P. infectoria Fuck., P. islandica Johans. and P. vagans Niessl., seven species of Drechslera including D. biseptata (Sacc. & Roum.) Richardson & Laser, D. dactylidis Shoemaker and D. sorokiniana (Sacc.) Subram. & Jain, seven species of Fusarium including F. avenaceum (Corda ex Fr.) Sacc., F. graminearum Schwabe, F. poae (Peck) Wollenweber and F. tricinctum (Corda) Sacc., and six species of leptosphaerioid fungi. In addition, rare fungi worthy of mention are Scopinella solani (Zukal) Malloch found in seeds of Agropyron repens, Bromus inermis and Festuca rubra; Ryparobius polysporus (Karst.) Sacc. found in seeds of Melica nutans; Drechslera verticillata Shoemaker found in seeds of Sieglingia brocumbens; and Curvularia protuberata Nelson and Hodges found in seeds of Festuca rubra. The most common fungi were Alternaria tenuis Nees, Cladosporium cladosporioides (Fres.) de Vries, Arthrinium phaeospermun (Corda) M. B. Ellis and Epicoccum purpurascens Ehrenb. ex Schlecht.

The largest number of fungus species were found on Melica nutans (46), Alopecurus pratensis (39), and Agropyron repens (37), whereas the lowest number of fungi were found on Bromus hordeaceus (15), Poa palustris (15), Anthoxanthum odoratum (16) and Calamagrostis phragmitoides (16). In the exceptionally rainy summer of 1977 the species of fungi were found to be more numerous than in the dry summer of 1975.

Index words: Grass seed, seed-borne fungi, Drechslera, Fusarium, Phaeosphaeria, Pleospora, Scopinella solani.

# INTRODUCTION

Numerous wild grasses are common throughout Finland e.g. Calamagrostis phragmitoides, Deschampsia caespitosa, D. flexuosa, Festuca rubra and Poa annua. Some grasses are more frequent in the south than in the north e.g. Agropyron repens, Alopecurus pratensis, Festuca pratensis and Poa pratensis (Hultén 1971). In addition, about 35 % of the total

arable land of Finland, 2,6 million hectares, is covered by grass (Anon. 1974 c, 1977 c).

The most important cultivated grasses are Phleum pratense and Festuca pratensis. Dactylis glomerata are also abundant, as well as law grasses, mainly Agrostis tenuis, Festuca rubra and Poa pratensis (Joy 1980, Laitinen and Laurila 1980).

The leaf spot diseases of cultivated grasses and the fungi producing them have been studied during the past decade in Finland (Mäkelä 1971, 1972 a, 1972 b). Many other microfungi have also been examined, on both

wild and cultivated grasses (Koponen and Mäkelä 1975, Mäkelä 1977 a, 1977 b).

Information on the seed-borne fungi of grass seeds is limited to the study of cultivated grass in both Finland (Mäkelä 1972 b) and other countries (Mühle 1953, 1971, Noble and Richardson 1968, Tulloch and Leach 1972, Neergaard 1977).

The aim was to study the species of microfungi occurring on the seeds of the wild grasses, relationships between the different species, and the incidence of these fungi in relation to different grass species, different years and different localities.

# MATERIALS AND METHODS

Samples of c. 300 wild grass seed lots were collected from field edges, meadows, forests, seashores and swards throughout the country, mostly from southern Finland (Fig. 1). The material comprised 63 grass species belonging to 36 genera (Table 1). They were mostly gathered in Uusimaa, Tvärminne (63 samples), in South Häme, Hattula (32 samples) and in South Savo, Lappeenranta (30 samples).

The samples were collected mainly from the end of June to the end of September: 4.7—26. 9. 1974, 20. 6—26. 9. 1975, 27. 6—29. 9. 1976, and 4. 7—4. 10. 1977. The seeds were dried and stored carefully, and were obtained from the Botanical Gardens, University of Helsinki (Anon. 1974 a, 1975 a, 1976 a, 1977 a).

The names of collectors are abbreviated as follows:

Pentti Alanko = P. A.
Pirkko Einistö = P. E.
L. O. Ervi = L. E.
R. Fagersten = R. F.
Sinikka Grahn = S. G.
M. Haapasaari = M. H.
Olavi Hankkila = O. H.

Ilkka Kukkonen = I. K.
Ilkka Kytövuori = I. Ky.
Marjaleena Miettinen = M. M.
Annikki Palmén = A. P.
Ernst Palmén = E. P.
Pirjo Pirinen = P. P.
Stina Saarnijoki = S. S.
Mirja Siuriainen = M. S.
M. & P. Uotila = M. & P. U.
Pertti Uotila = P. U.
Yrjö Vasari = Y. V.
Seppo Vuokko = S. V.

The nomenclature of the vascular plants is according to Hylander (1953), with a few exceptions. The abbreviations for biological provinces are in accordance with Heikinheimo and Raatikainen (1971).

One or two hundred seeds were examined from each lot of seeds. Germination occurred in a Jacobsen's incubator on moist filter paper under a perforated glass dome. The average temperature was  $+20^{\circ}$ C. The temperature at night was often a few degrees lower than the daytime temperature. The tests were exposed to light in daytime but

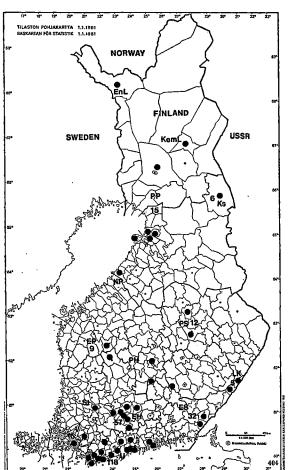


Fig. 1. The origin of the seed samples of grasses by locality. Number of seed lots by biological province.

were in the dark at night (cf. Anon. 1966, Tempe 1963).

The germination and fungi of the seed lots were examined simultaneously twice, 6 and 14 (21, 28) days after the beginning of germination. In addition, examinations of a more general nature were carried out about a month after the beginning of germination. During this culture period the fungi were examined a few times with a stereomicroscope. In addition, a light microscope was used for measurements and microphotographs. Slides were prepared with lactic

Table 1. Place of origin and number of the

Agropyron caninum (L.) PB.

A. repens (L.) PB.

Grass

Agrostis canina L. A. gigantea Roth A. stolofinera L. A. tenuis Sibth. A. vinealis Schreber Alopecurus aequalis Sobol. A. geniculatus L. A. pratensis L. Anthoxanthum odoratum L. Apera spica-venti (L.) PB. Arrhenatherum elatius (L.) J. & C. Presl Avena fatua L. Avenula pubescens (Hudson) Dumort. Bromus hordeaceus L. B. inermis Leysser Calamagrostis arundinacea (L.) Roth C. canescens (Weber ex Wiggers) Roth C. epigejos (L.) Roth C. phragmitoides Hartman C. stricta (Timm) Koeler C. varia (Schrad) Host Dactylis glomerata L. Deschampsia bottnica (Wahlenb.) Trin. D. caespitosa (L.) PB. D. flexuosa (L.) Trin. Elymus arenarius L. Festuca arundinacea Schreber F. ovina L. F. polesica Zapat. F. pratensis Hudson F. rubra L. F. trachyphylla (Hackel) Krajina Glyseria fluitans (L.) R.Br. G. lithuanica (Górski) Lindman G. maxima (Hartman) Holmberg Hierocloë australis (Schrader) Roemer & Schultes H. hirta (Schrank) Borbás H. odorata (L.) PB. Holcus lanatus L. H. mollis L. Melica nutans L. Milium effusum L. Molinia caerulea (L.) Moench Nardus stricta L. Phalaris arundinacea L. Phleum commutatum Gaud. P. phleoides (L.) Karsten P. pratense L. Phragmites australis (Cav.) Trin. ex Steudel Poa annua L. P. compressa L. P. nemoralis L. P. palustris L. P. pratensis L. P. trivialis L. Puccinellia capillaris (Liljebl.) Jansen P. distans (L.) Parl. Roegneria borealis (Turcz) Nevski Sieglingia decumbens (L.) Bernh. Trisetum spicatum (L.) Richter Total 63 Grass species

grass seed lots examined. The samples were collected in 1974—1977.

	1974	v)	1975	<u>.</u>	1976	- vs	1977	o	Total in 1974—1977
No. of lots examined	Place of origin	of lots	Place of origin	of lots	Place of origin	f lots	Place of origin	f lots	Place of origin
o. of						o, of		o. of	1
ĕ×		No.		No.	·	No.		No.	
2	U, Ks	3	U, EH, KemL	1	PP	1	PH	7	U, EH, PH, PP, Ks, KemL
2	υ	2	ΰ	$\frac{2}{1}$	U, EH ES	$\frac{1}{2}$	ES ES, EP	$\frac{7}{3}$	U, EH, ES ES, EP
1	ŭ	1	U	1	υ		•	3	U
1 1	U U	1 1	U PH	2	U, EK	2	U, EH	2 6	U U, EH, EK, PH
1	υ	1	υ`	2	EH, ES	1 1	V EH	1 5	V U, EH, ES
1	· <b>ʊ</b>	_						1	บั
3 1	V, U, EH U	2 1	U, PH U	5 1	U, EH, ES, PP ES	6 3	U, EH, ES, EP U	16 6	V, U, EH, ES, EP, PH, PP U, ES
1	Ŭ	•	ŤŤ	1	ES	-		2	U, ES
1	EH	1	Ū	1	Ŭ			2 1	U EH
1	υ	1	U U	1	U	$\frac{2}{1}$	U, EH U	4 4	U, EH U
1 1	U EH	1	EH	1	ES	2	U, ES	5	U, EH, ES
2	U, ES	1	V PH	2	ES, PS	5 2	V, U, EP EH, ES	$\frac{10}{3}$	V, U, ES, EP, PS EH, ES, PH
2	U, ES	1	Ü	2	U, PS	4	U, EH, EP, PS	9	U, EH, ES, EP, PS
1	Ks	$\frac{1}{2}$	U U, EH	1 3	PS ES, PP	2	EP, PP	5 8	U, EP, PS, PP, Ks U, EH, ES, LK, PP
3 1	U, LK, PP		•					1	
2	EH	2	U, EH	4 1	U, EH, ES, PS PP	3	U, EH	11	U, EH, ES, PS PP
4	U, EH, LK, EP	2	U, PH	4	U, LK, PS, PP	8	U, EH, ES, PS, PP	18	U, EH, ES, LK, EP, PS, PP
1	U TI DD	$\frac{1}{2}$	U U	$\frac{2}{1}$	EK, PP PP	$\frac{4}{2}$	V, EP, PS U	8 7	V, U, EK, EP, PS, PP U, PP
$\frac{2}{1}$	U, PP U	1	$\breve{\mathtt{v}}$	î	Ŭ .	2	Ŭ	5	Ŭ
1	Ω,	2 1	U, PH U			3	V, EH, ES	$\frac{6}{2}$	U, EH, ES, PH U
· 1	U EH, LK	1	EH	3	U, EH, ES	3	U, EH, ES	9	U, EH, ES, LK
1	U	1	U	1	ES	2 1	ES, PP U	5 1	U, ES, PP U
1	υ	1	U		Da			2	U
		1	EH	2	PS	1	EH EH	4 1	EH, PS EH
1	U	ก	II DII	1 1	U OO		TATE	2	\U ET DD
2	U, Ks	2	U, EH	1	PP	1	EH	$\frac{4}{2}$	U, EH, PP U, Ks
	·	1	ŭ					1	Ū
1 4	U U, EH, Ks	$\frac{1}{2}$	U U, EH	3	U, EH, ES	6	U V, U, EH, PS	3 15	U V, U, EH, ES, PS, Ks
1	U	1	<b>บ</b> ์	1	EH	1	PS	4	U, EH, PS
1 1	U U	$\frac{2}{1}$	U U	1	Ŭ	3 1	U, ES, EP ES	7 3	U, ES, EP U, ES
4	U, EH, ES, LK	2	U, EH	3	EH, ES, KP	3	U, EH, ES	12	U, EH, ES, LK, KP
2	Ks, EnL			1	EH			2 1	Ks, EnL EH
2	U, PH	3	U, EH, PH	3 1	U, EK, EH ES	4 1	EH, ES, EP ES	$^{12}_2$	U, EK, EH, ES, EP, PH ES
		1	v	T	LS	1	ь	1	V
1	Ŭ	1	U	1	ES	$\frac{1}{2}$	ES V, U	3 4	U, ES V, U
1 1	U EH	1 1		1	PP	$\frac{2}{2}$	EH	5	U, EH, PP
3	U	2	EH, PH	1	EH	1	υ	7 2	U, EH, PH U
1 1		1 1						2	U
						1	EH	1 1	EH
1		1	บ					1	Ŭ
_1						_		, 1	· <del></del>
72	-	60		64		93		289	135

Table 2. Frequency of the seed borne fungi, percentage of infected seeds on different grasses examined in 1974—1977.

						Fı	equency	of fun	gi, perce	entage	of in	fected s	eeds by	grass	species									· <u> </u>			Ye	ar	
Fungi	Agropyron caninum	A. repens	Agrostis tenuis	Alopecurus aequalis	A. pratensis	Anthoxanthum odoratum	Bromus hordeaceus	B. inermis	Calamagrostis arundinacea	C. epigejos	C. phragmitoides	Dactylis glomerata	Deschampsia caespitosa	D. flexuosa	Elymus arenarius	Festuca arundinacea	F. pratensis	F. rubra	Melica nutans	Milium effusum	Molinia caerulea	Phalaris arundinacea	Phleum pratense	Poa palustris	P. pratensis	1974	975	976	776
No. of seed lots Germination <sup>0</sup> / <sub>0</sub>	8 66	7 17	6 30	5 65	16 14	6 49	4 90	5 7	10 3	9 2	5 0,8	11 18	18 46	8 32	7 12	5 62	9 45	7 74	15 26	4 36	7 3	12 13	12 94	5 52	7 55	46 40	38 46	49 34	72 26
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29
Myxomycetes Physarales Didymium spp. Zygomycotina Mucorales Mucor spp.	0,2	2		0.5	0,5			0,5	0,5	0,3	0.3	0,7 :	+	0,1	0,1	1	1	5	0,2	0,1	0,1	0,3	0,3			0,3	+	2	1
Rhizopus nigricans Ehrenb, Ascomycotina Pyrenomycetes	0,3	1	0,3	0,5 2	0,1	1	9		0,5	0,3 0,1	0,3 1	1	0,6 0,3	0,1	0,5 18	0,3	$^{4}_{0,2}$	0,3 4	2 11	4	0,5 0,3	1	0,1 +	1	0,3 1	$0,2 \\ 0,4$	1 3	1 6	+ 0,4
Sphaeriales Ceratocystis sp. Chaetomium spp. Claviceps purpurea (Fr.) Tul. Gibberella zeae (Schn.) Petch. Monographella nivalis (Rehm) E. Müller	0,6	6 0,4	1	5	0,1 0,1 0,1		0,3		0,1	3	0,5	0,1	4		0,3				0,3			0,3				0,1 1 0,3	1 0,1	0,2	1 0,3 + +
Podospora spp. Scopinella solani (Zukal) Mallach Sordaria spp. Sporormia intermedia Auersw. Loculoascomycetes		•		+				1		-	-				1			3	0,3 0,2							0,1 0,5	+	+ 0,1	0,6 0,2
Pleosporales Leptosphaeria spp. Pleospora spp. Pyrenophora sp.	0,1	0,4 2	0,5 0,1		1 0,4			0,3	Ó,1			0,3	0,2		0,5 0,3	1 0,4 0,5	0,6 0,1	1	, 2	1	$^{0,1}_{0,2}$				1	0,4 +	+	0,1 0,2	1 0,6
Discomycetes Rybarobius polysporus (Karst.) Sacc. Basidiomycotina																-,-	-	•	+					,		+			0,1
Holobasidiomycetidae Rhizoctonia solani Kühn Deuteromycotina													0,2		r.				0,1			0,1							+
Hyphomyeetes Acremoniella atra (Corda) Sacc. Acremonium spp. Alternaria tenuis Nees Arthrinium phaeospermum (Corda) M. B. Ellis Arthrobotrys suberba Corda Aspergillus spp. Botrytis cinerea Pers. ex Fr. Cladosporium spp. Curvularia protuberata Nelson & Hodges Epicoccum purpurascens Ehrenb. ex Schlecht Fusarium spp. Fusarium avenaceum (Corda ex Fr.) Sacc. Fusarium poae (Peck) Wollenweber Fusarium semitectum Berk. & Rav. Fusarium tricinctum (Corda) Sacc. Fusarium sp. Gonathobotrys simplex Corda	2 60 0,3 1 53 13 0,3 0,3	2 93 1 0,1 33 56 0,6 1 0,5 0,1 0,3 5	3 0,1 16 7 30 5 0,4 0,1 0,1	31 1 0,3 26 6 0,3	3 82 0,2 0,2 0,4 15 22 2 0,6 0,2 0,6 0,3 0,6 0,1	1 48 1 1 15 5 0,1	0,3 48 1 1 8 4 0,5	2 79 0,1 25 31 0,3 1 0,1 0,1 0,3 5	0,7 25 0,3 0,1 0,4 28 10 0,5	4 35 1 0,1 1 32 19 0,2	0,5 14 0,1 2 0,1 27 1	6 65 4 0,5 41 23 3 1	0,2 24 4 0,3 + 21 20 1 0,1	1 20 4 0,7 0,1 35 6 3 0,4 0,6 1	4 92 0,5 3 1 30 23 4 0,1	0,1 85 0,3 28 10 1	0,5 71 1 0,3 20 16 4 1 0,3 0,3 0,3	2 65 0,1 0,1 17 0,2 24 3 0,1 0,6	2 42 0,2 1 1 39 17 1 0,1 0,1 0,1 0,2	0,5 6 0,3 0,3 0,1 22 3 2	0,1 50 3 29 14 1 0,3 0,2	0,1 1 51 0,4 1 36 12 0,1	0,6 50 1 0,3 0,1 39 15 2 0,5 0,6	6 19 1 38 8 1	0,6 0,4 57 1 0,6 9 26 1	+ 2 43 1 0,7 0,1 17 16 0,6 0,2 +	0,4 0,4 42 + 1 0,1 30 19 0,1	+ 0,4 56 2 1 0,6 11 14 0,4 0,2 0,1 0,1 0,6	0,2 3 59 2 0,1 0,2 0,7 54 0,2 13 4 0,5 0,3 0,4 1 1

Table 2. (cont.)

								_		<del></del>	—:	entage (	of infer	ted sec	de hv ~	mace or	———								- A				
								Freque	ncy of f	ungi,		einage .	or illied	ieu see	us by g	rass spe	ecies								,		Y	ear	
Fungi	Agropyron caninum	A. repens	Agrostis tenuis	Alopecurus aequalis	A. pratensis	Anthoxanthum odoratum	Bromus hordeaceus	B. inermis	Calamagrostis arundinacea	C. epigejos	C. phragmitoides	Dactylis glomerata	Deschampsia caespitosa	D. flexuosa	Elymus arenarius	Festuca arundinacea	F. pratensis	F. rubra	Melica nutans	Milium effusum	Molinia caerulea	Phalaris arundinacea	Phleum pratense	Poa palustris	P. pratensis	1974	1975	1976	1977
No. of seed lots Germination <sup>0</sup> / <sub>0</sub>	8 66	7 17	6 30	5 65	16 14	6 49	4 90	5 7	10 3	9 2	5 0,8	11 18	18 46	8 32	7 12	5 62	9 45	7 74	15 26	´ 4 36	7 3	12 13	12 94	5 52	7 55	46 40	38 46	49 34	72 26
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29
Graphium sp. Helminthosporium spp. H. biforme Mason & Hughes Oedocephalum glomerosum (Bull.) Sacc. Oidiodendron cerealis (Thüm.) Barron	2 0,3 0,3	0,4 0,4 3	0,5	0,3	0,1 0,1 0,5	2 2		2	0,2 0,1	0,1 0,3	!	0,1 4	0,3	0,1 5 2	0,3 4 0,3	0,5 0,4	0,2 0,4	2 0,5	0,1 0,1 0,6 0,1	0,3	0,5	. 1	0,3	1	0,5 0,6	0,8 0,4 0,1	0,2 0,6 0,4	0,1 0,3 0,5 0,4	+ 1 0,4 0,1 +
Ostracoderma state of Peziza ostracoderma Korf Penicillium spp. Periconia digitata (Cooke) Sacc. Rhinocladiella spp. Scopulariopsis brevicaulis (Sacc.) Bain.	17 24 0,3 4	29 4 1	0,4	2 3	8 2 0,3	1·0 10	15 19	16 2 3	4 5	<b>4</b>	3	6 0,6 0,1 0,1	1 2 0,1 +	4 4 0,2	10 5 4 0,3	3 1 0,5	12 21 1	3 1 0,6	0,1 9 0,1 0,3	11 3	1 7 0,2 0,1	2 3	8 0,4 0,1	0,3 3	2 4	0,6 5	6	17 5 0,1 +	+ 11 6 0,1 2
Stachybotrys atra Corda Stemphylium botryosum Simmons Tricocladium asperum Harz Trichoderma viride Pers. ex Fr. Trichothecium roseum Link ex Fr. Ulocladium consortiale (Thüm.) Simmons Verticillium spp. Volutella ciliata (Alb. et Schn.) Fr.	1 2 1	2 0,1 2 2	0,5	0,3	0,1 0,1 1 0,1	0,2	1	0,3 3 0,5	0,1 0,1 0,6	+ 1 0,1	. 0,2	0,5 0,4 5 3	0,3 0,1 0,1 +	0,3	0,5 0,1 5 4 0,3	0,4 0,5 0,1	1 10	2	0,1 0,1 0,4 1		0,3 0,1	0,1 0,3 1	1	0,3	0,1 0,1 0,3	+ + 0,3 + +	+ + 0,5 +	0,4 0,1 + 0,5 2	0,5 0,1 + 1 0,4 0,5
Coelomycetes Melanconiales Colletotrichum graminicola (Ces.) Wils. Truncatella truncata (Lév.) Stey Sphaeropsidales Ascochyta spp.		0,2		0,5				÷			· ·		+	0,1			0,1		1 0,3	0,3	0,1		0,1	, t		++		4	1 0,2
Camarosportum sp. Coniothyrium spp. Dinemasprorium gramineum Lev. Diplodia sp. Hendersonia spp. H. crastophila Sacc.	0,3	0,0	0,1	0,3	0,1	0,3		1	2				+ + 0,2	0,4	0,3	0,1	0,2 0,1	0,1	0,1 1 0,7 0,1	0,3	0,3 0,1		0,1		1	0,5 0,2	+ + +	+ + 0,2	0,1 0,2 0,2 +
H. culmicola Sacc. Phaeoseptoria festucae Sprag. Phoma spp. Septoria spp. Stagonospora sp. Unidentified fungi	.5 0.6	1	0,4	1	0,1 6 0,1 0,4	1	18 0,5	6 0,5	0,5 1 0,6	0,4 0,4	0,1 0,5	0,1 0,3	0,1 0,3 2	0,1 2 8	0,3 0,3	0,3	0,3 1	0.5	0,1 5	0,3	0,5	0,1 1	0,1	0,6 0,3	0,3 2 0,3	0,4 0,6	3 +	+ 4	0,2 + 0,3 1 + +
Bacteria Actinomycetales Streptomycetes spp.	4	3	0,4		0,3			1	0,3	0,1		2		0,1	0,5	0,1	7	1	3	<u>.</u>	1	0,2	1	1	0,5	0,1	1	3	1
Total no. of fungi Total disease percentage	27 196	37 268	21 68	19 89	39 151	16 99	15 126	25 181	26 82	22 104	16 52	29 177	33 83 <sup>.</sup>	25 98	33 214	25 136	31 182	27 138	46 158	19 55	27 112	23 112	23 121	15 82	24 110	43 94	33 111	43. 133	62 175

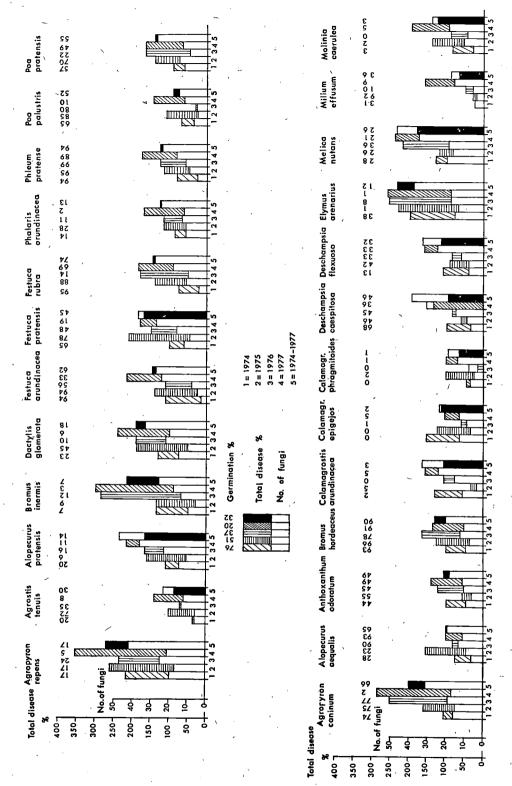


Fig. 2. Occurrence of the seed borne fungi, percentage on different grasses examined in 1974—1977.

acid, and these also were measured and photographed. The films are in the keeping of the author.

The degree of contamination of the seed was not observed in this study. The results (Table 2) show only the occurrence of a pathogen in the seed, not its quantity. The frequency of occurrence of the fungi in the seed lots is an average according to the grass and years examined. The total disease figure (per cent) expresses the sum of the occurrence of all the fungi on the grass species.

# Weather conditions

The summer of 1974 was exceptional by rainy. The summer of 1975 was very warm and dry. Long dry periods occurred particularly in Kymenlaakso and South Häme. The growing season of 1976 as a whole was cool and somewhat drier than normal. The summer of 1977 was in general cool and rainy (Anon. 1974 b, 1975 b, 1976 b, 1977 b).

## RESULTS

The species of fungi encountered in the seed lots varied according to the grass species, the year and the habitat of the plant (Table 2, Fig. 2).

# Effect of grass species

The number of fungi varied greatly in the samples of grass examined. The seeds of Melica nutans contained the largest number of fungi (total 46, range 7-22 fungi per lot). The corresponding figure for Alopecurus pratensis was 39 fungi (range 4-23) and for Agropyron repens 37 fungi (range 14-24). On the other hand, the seeds of Poa palustris contained the lowest number of fungi (total 15, range 5-12 fungi per lot). The corresponding figure for Bromus hordeaceus was also 15 fungi (range 8-12), for Calamagrostis phragmitoides 16 fungi (range 5-11) and for Anthoxanthum odoratum 16 fungi (range 5—12). The largest number (24 fungi per individual seed lot) was found on Agropyron repens and the lowest number (3 fungi) on Festuca rubra and on Phleum pratense.

The number of fungi corresponded with the total disease in many cases. The total disease percentage was high on, e.g. Agropyron repens, A. caninum and Elymus arenarius, and it was low on, e.g., Calamagrostis phragmitoides, Milium effusum, Agrostis tenuis and Poa palustris.

# Effect of the year

There were great annual variations in the number of fungi and in the total disease. Least fungi were found in the warm and dry summer of 1975 and most in the cool and rainy summer of 1977.

## Germination of the seeds

The germination was dependent on the grass species and the year. The highest germination percentage (average 94 %) was achieved by the seeds of *Phleum pratense*. The seeds of *Bromus hordeaceus*, *Festuca* and *Poa* species also germinated moderately well. On the other hand, the seeds of *Molinia caerulea*, *Calamagrostis* species and *Bromus inermis* germinated poorly (average only 1—7 %). There was also a high annual variation in the germination of the seeds. The percentage germination was highest in 1975;

Table 3. The systematic distribution of the fungi on the grass seed lots examined.

		Fu	ngi
Fungi		No. of genera	No. of species
Myxomycetes		1	4
Zygomycotina			
Mucorales		2	2
Ascomycotina			
Pyrenomycetes			
Sphaeriales	500	9	12
Loculoascomycets			
Pleosporales		3	11
Discomycetes		2	2
Basidiomycotina			
Holobasidiomycetidae		1	1
Deuteromycotina			
Hyphomycetes		29	48
Coelomycetes			
Melanconiales		2	2
Sphaeropsidales		10	11
	Total	59	93
Bacteria			
Actinomycetales		1	

<sup>1</sup> an approximate estimate

twenty-five grass species gave an average of  $46\,\%$  (range  $0-96\,\%$ ), whereas in the unfavourable summer of 1977 the seeds of the same grass species germinated with averages of  $26\,\%$  (range  $0-93\,\%$ ).

# Species of fungi

The total number of fungal species found numbered over 90, belonging to 60 genera (Table 3). Nearly 70 % of the fungal species belonged to the class *Deuteromycotina* and of these the majority were members of the order *Hyphomycetes*. Sphaeropsidales was also well represented. Approximately 25 % of the fungi belonged to the class Ascomycotina, most of them being Pyrenomycetes.

#### Muxomucetes

Didymium difforme (Pers.) S. F. Gray was the most common species of the class. Found in 19 grass species in 12 localities throughout the country (U, EH, ES, EP, PS, PP). Com-

mon on grains (Härkönen and Koponen 1978), infrequent on ripe cereals in Finland (Mäkelä and Mäki 1980).

Didymium iridis (Ditmar) Fr.

Three specimens: On Alopecurus pratensis (U: Helsinki), Arrhenatherum elatus (U: Tenhola), Festuca rubra (ES: Lappeenranta). Occasionally on grains (Härkönen and Koponen 1978), and on ripe cereals in Finland (Mäkelä and Mäki 1980).

Some unidentified species of fungi belonging to the class *Myxomycetes* were also found.

Zygomycotina, Mucorales

The *Mucor* and *Rhizopus* fungi were more common in the dry summers of 1975 and 1976 than in the rainy summers. *R. nigricans* was usually found in the seed of *Elymus arenarius* and *Melica nutans*.

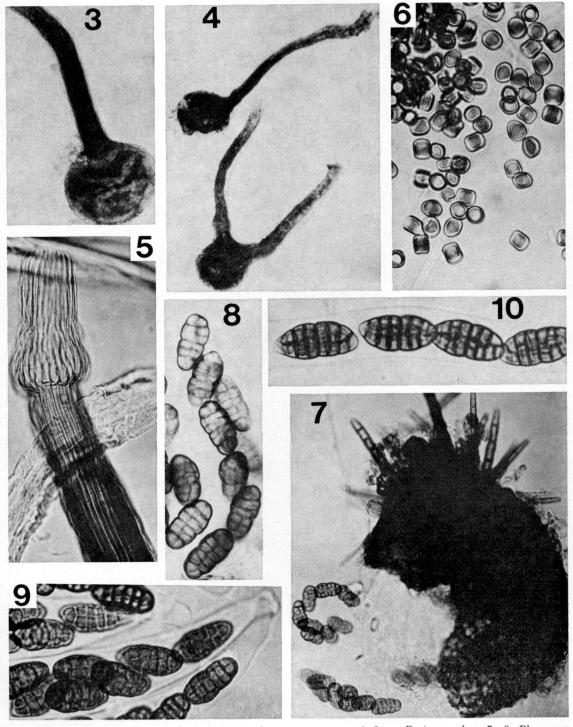
Ascomycotina, Pyrenomycetes Ceratocystis sp. was found only in the seed of Agropyron repens (U: Tvärminne) in 1974 and 1975.

Claviceps purpurea (Fr.) Tul. was found on eleven grass species throughout the country; most common on Alopecurus aequatilis, Deschampsia caespitosa and Calamagrostis epigejos, particularly in the rainy summers of 1974 and 1977. Very common on grasses in Fennoscandia (Eriksson 1967 b). Cosmopolitan (Sprague 1950, Neergaard 1977).

Podospora curvula (de Bary) Niessl Three specimens: on Deschampsia flexuosa (U: Tvärminne), and Melica nutans (U: Tvärminne, ES: Lappeenranta).

Sordaria fimicola (Rob.) Ces. & de Not One specimen: on Elymus arenarius (U: Tvärminne). Found in one seed lot of Dactylis glomerata from Finland (Mäkelä 1972 b). Cosmopolitan (Domsch and Gams 1970).

Scopinella solani (Zukal) Malloch (Figs. 3—6)



Figs. 3—10. 3—6: Scopinella solani, 3, 5: on Agropyron repens, 4, 6: on Festuca rubra. 7—9: Pleospora infectoria, 7: on Festuca arundinacea, 8: on Alopecurus pratensis, 9: on Deschampsia caespitosa. 10: Pleospora islandica on Deschampsia caespitosa.

Material: 3, 5: U, Tvärminne, 3: 14.8.1974 A.P., 5: 14.8.1975 A.P. 4, 6: ES, Lappeenranta 28.8.1977 S.V. 7: U, Helsinki 4.10.1977 S.V. 8, 9: EH, Hattula, 8: 21.8.1977 P.U., 9: 17.9.1977 M.M. 3: X 200. 4: X 100. 5: X 600. 6, 7, 9, 10: X 900. 7: X 400.

Five specimens: on Agropyron repens (U: Tvärminne, 14. 8. 1974 A. P., 14. 8. 1975 A. P.). On Bromus inermis (ES: Lappeenranta, 6. 9. 1976 S. V., 28. 8. 1977 S. V.). On Festuca rubra (ES: Lappeenranta, 28. 8. 1977 S. V.). Ascocarp red-brown (122—) 165 (—202)  $\mu$ m diameter with a long neck (185—) 493 (—1000) X (32—) 48 (—92  $\mu$ m. Ascospores (3,4—) 5,5. (—6,9) X (3,4—) 5,2. (—6,9)  $\mu$ m. Very rarely mentioned (Malloch 1976).

Loculoascomycetes, Pleosporales
Paraphaeosphaeria michotii (Westend) O.
Erikss. Found on six grass species, including
Agrostis tenuis (EH), Deschampsia caespitosa
(U), D. flexuosa (EP), Festuca ovina (V),
Calamagrostis arundinacea (V), and F. trachyphylla (U). Reported on nine grass species
in Finland (Koponen and Mäkelä 1975).
Found sporadically in Fennoscandia (Eriksson
1967 b).

Phaeosphaeria eustoma (Fuck.) L. Holm Three specimens: on Agrostis canina (ES), Alopecurus pratensis (ES), and Deschampsia flexuosa (EP). Observed on 16 grass species throughout Finland (Koponen and Mäkelä 1975). The fungus is rather common on grasses in Fennoscandia (Eriksson 1967 b).

Phaeosphaeria herpotrichoides (De Not.) L. Holm

Found on nine grass species (from EH, ES, EP, Ks), including *Agrostis canina* (ES), *Festuca pratensis* (EH) and *Poa compressa* (ES); recorded for the first time from Finland. Observed on 34 grass species, very common throughout Finland (Koponen and Mäkelä 1975). Very common in Fennoscandia (Eriksson 1967 b).

Phaeosphaeria microscopica (P. Karst.) O. Erikss.

Eight specimens found on six grass species: Alopecurus pratensis (EP), Avenula pubescens (EH), Calamagrostis arundinacea (EP), Deschampsia caespitosa (EP, PS, PP), and Glyceria maxima (EH). Found on 26 grass

species collected from throughout Finland (Koponen and Mäkelä 1975). Common on grasses in Fennoscandia (Eriksson 1967 b).

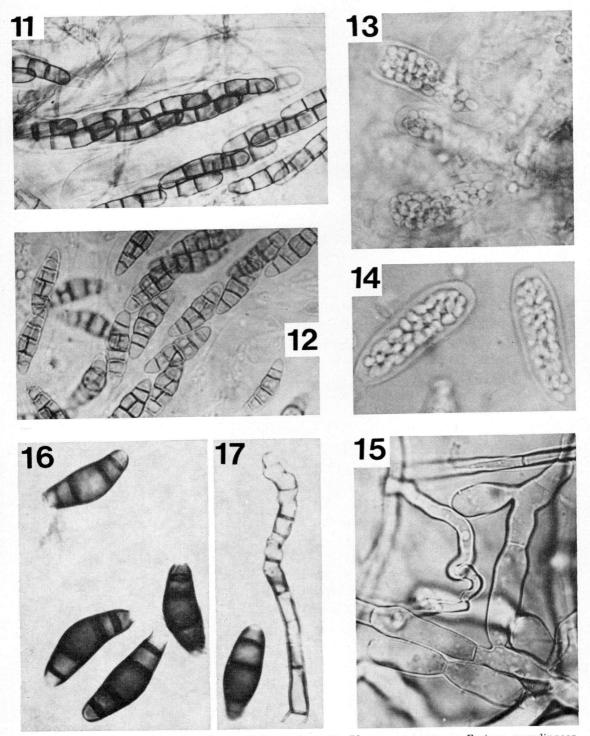
Phaesphaeria nodorum (E. Müll.) Hedjaroude

Found on thirteen grass species (from U, EH, ES, EP, PS); only in 1977: Agrostis canina (ES), A. tenuis (EH), Avenula pubescens (EH), Bromus hordeaceus (U), B. inermis (ES), Calamagrostis phragmitoides (EP), Deschampsia flexuosa (EP) and Festuca arundinacea (U) are recorded for the first time from Finland. Reported on 22 grass species in southern Finland (Koponen and Mäkelä 1975).

Phaeophaeria vagans (Niessl) O. Erikss. Found on fifteen grass species (from U, EH, ES, EP, PH, PS); only in 1977: Agropyron caninum (PH), Agrostis canina (ES), Calamagrostis arundinacea (EP), C. canescens (ES), Deschampsia flexuosa (EP), Festuca arundinacea (U), Hierochloë hirta (EH) and Molinia caerulea (U) are recorded for the first time from Finland. Observed to be very common on grasses throughout Finland (Koponen and Mäkelä 1975). Very common in Fennoscandia (Eriksson 1967 b).

Pleospora infectoria Fuck (Figs. 7—9) Found on six grass species between August 4 and October 4 in 1977: on Agropyron repens (ES: Lappeenranta), Agrostis tenuis (U: Helsinki), Alopecurus pratensis (U: Riihimäki, EH: Hattula), Deschampsia caespitosa (EH: Hattula, 2. exx, PP: Rovaniemi), Elymus arenarius (ES: Lappeenranta) and Festuca arundinacea (U: Helsinki). Pseudothecia (106—) 193 (—315)  $\mu$ m with setose. Asci (70—) 81 (—104) X (14—) 17 (—23)  $\mu$ m. Ascospores (50 spores) (16—) 19 (—23) X (7—) 9.3 (—12)  $\mu$ m. The fungus is common in southern Fennoscandia (Eriksson 1967 a).

Pleospora islandica Johans. (Fig. 10) Two specimens: on *Deschampsia caespitosa* (EH: Hauho, 9. 8. 1977 M. M., PS: Lapinlahti, 3. 8. 1977 M. M.). Pseudothecia 405—460 μm



Figs. 11—17. 11: Sporomia intermedia on Melica nutans. 12: Pleospora vagans on Festuca arundinacea. 13, 14: Ryparobius polysporus on Melica nutans. 15: Rhizoctonia solani on Melica nutans. 16, 17: Curvularia protuberata on Festuca rubra.

Material: 11: U, Tvärminne 15. 7. 1974 A.P. 12: U, Helsinki 11. 9. 1977 P.U. 13, 14: Ks. Kuusamo 23. 8. 1974 S.S. 15: U, Porvoo rural district 17. 7. 1977 A.P. 16, 17: ES, Lappeenranta 26. 9. 1976 S.V. 11: X 500. 12, 15—17: X 700. 13, 14: 900.

in diameter. Asci (150—) 159 (—182) X (14—) 19 (—23)  $\mu$ m. Ascospores (23—) 25 (—28) X (11,5—) 11,8 (—13,8)  $\mu$ m. Very common in the arctic regions of Fennoscandia (Eriksson 1967 a).

Pleospora vagans Niessl. (Fig. 12) Found on nine grass species: on Agropyron caninum (U: Tvärminne, 6. 9. 1975 E. P.), Agrostis gigantea (U: Tvärminne, 15. 8. 1974 A. P.), Agrostis tenuis (EH: Hattula, 19. 8. 1977 P. U.), Alopecurus aequalis (U: Porvoo, 25. 9. 1974 A. P.), Bromus inermis (U: Helsinki, 29. 9. 1977 S. V.), Calamagrostis arundinacea (U: Espoo, 20. 9. 1977 S. V.), Deschampsia flexuosa (PS: Lapinlahti, 2. 8. 1977 M. M.), Festuca arundinacea (U: Helsinki, 11. 9. 1977 P. U., 4. 10. 1977 S. V.), and Poa pratensis (U: Tvärminne, 26. 9. 1974 A. P.). Pseudothecia 138—230 µm across, ascospores (25—) 30 (—35) X (7—) 8,6. (-11.5) um. Found on Triticum aestivum (U: Siuntio) (Mäkelä and Mäki 1980). The fungus is very common on grasses in Fennoscandia (Eriksson 1967 a).

# Discomycetes

Ryparobius polysporus (Karst.) Sacc. (Figs. 13—14)

One specimen: on *Melica nutans* (Ks: Kuusamo, 23. 8. 1974 S. S.). Apothecia 72—90  $\mu$ m across, asci 47  $\times$  13,8  $\mu$ m, ascospores 2,7.  $\times$  2,3  $\mu$ m (cf. Kimbrough 1965). Found in Finland (Karsten 1871 a, 1871 b, 1885). Very rarely mentioned (Domsch and Gams 1970).

Basidiomycotina, Holobasidiomycetidae The basidiomycetes were extremely scarce in the samples examined.

Rhizoctonia solani Kühn found on three specimens (Fig. 15): Deschampsia caespitosa (? 1977), Melica nutans (U: Porvoo, 17. 7. 1977 A. P.), and Phalaris arundinacea (EH: Hattula, 19. 8. 1977 P. U.).

# Deuteromycotina

The fungi of this class were by far the most Arthrinium phaeospermum, Epicoccum pur-

Table 4. Fusarium species on the seed lots and observations by provinces.

	жап	F. ave	naceum
Grass	No. of lots examined	No. of lots	Recorded from
Agropyron caninum	7		<u> </u>
A. repens	7	2	EH, ES
Agrostis canina	3	1.	ES
A. tenuis	6	2	U
Alopecurus aequalis	5		
A. pratensis	16	4	U, ES, EP
Anthoxanthum odoratum	6		
Avenula pubescens	4	<b>2</b>	U, EH
Bromus hordeaceus	4		÷.
B. inermis	5	2	EH, ES
Calamagrostis arundinacea	10	•	•
C. epigeios	9		
C. varia	1	1	
Dactylis glomerata	11	3	U, EH, PS
Deschampsia caespitosa	18	1	Ŭ
D. flexuosa	8	1	v ·
Elymus arenarius	7	.1	PP
Festuca ovina	6	1	ES
F. pratensis	9	3	U, EH, ES
F. rubra	5	1	ES
F.trachyphylla	1.		
Glyceria lithuanica	4	1	EH
G. maxima	1		
Hierochloë hirta	4		
Holcus lanatus	1	1	U
Melica nutans	15	1	EH
Milium effusum	4		
Molinia caerulea	7		
Nardus stricta	3		
Phalaris arundinaceae	12		
Phleum commutatum	2		
P. pratense	$\frac{12}{2}$		
Phragmites australis	3	1	THE .
Poa compressa	ა 4	T	ES
P. nemoralis P. palustris	5		
P. pratensis	7		
Sieglingia decumbens	1	1	U
——————————————————————————————————————			
38 Grass species Total	235	30	
Percentage of lots infected	82,7	10,6	
	(	•	1077
·	1	18	1977
Found in 1974—1977	{	$\frac{4}{2}$	1976
	1	Z	1975

numerous in this study. Most frequent among them were Alternaria tenuis, Cladosporium cladosporioides (Fres.) de Vries, Arthrinium phaeospermum. Epicoccum pur-

	ulmo- ım	F. 9		F. 1			poae	F.	semi- ectum	F. $t$	s by biological ricinc- tum	Fus	sarium spp.	lots
No. of lots	Recorded from	No. of lots	Recorded	No. of lots	Recorded from	No. of lots	Recorded from	No. of lots	Recorded from	No. of lots	Recorded from	No. of lots	Recorded from	Total no. of lots
	•					1	PP ES	2	EH, ES	1	ES	1 2 3	PH ES, EH ES	2 3 3
		1	EH			1 2	U U, PP	3	EH, ES, EP	3	EH, EP, PS	2 1 6	U, EK EH U, EH, PP	4 1 9
1	υ							1 1	EH U			1 1 1	U U U	1 2 2
								1	EP	1	υ	1 3 1	ES V, U, EK EP	3 4 1
1	υ.	2	υ					1	<b>v</b>	2 1 2	U U V, EP	3 6 3 3	U, EH U, EH, ES, EP, PP PP V, EP, PP	1 6 6 3
		1 1	V ES	1	v	1	EH	2 1	U, EH ES	3	u, eh, es	4 4 2	V, U, EH, ES U, EH, ES ES, PP	3 4 5 2
			<b>Y</b> 3Y7			1	EH U		,	9	***	1 1 1 1 5	U EH EH U U	1 1 2 1 7
		1	EH		·	1 1 1 1	Ks PS U ES	1	EH	2	V ES	1 3 1	V, U, EH, PS PS U, ES, EP ES	1 3 1
		1	ES			2	EH	1	ES	2	EH, ES	1 1 4 1	EH, ES ES	1 1 4 1
1	v	•	110					1 2 1	ES V, U EH	1	ES	1 2 2	ES V, U EH	$\begin{array}{c} \cdot & 1 \\ 2 \\ 2 \end{array}$
										1	U	2	EH	2 1
3 1,1 2	1077	7 2,5 7	1977	1 0,3		14 4,9	1000	19 6,7		20 7,0	1055	77 27,1	1005	171 60,2
1	1977 1974		1911	1	1976	6 5 2 1	1977 1976 1975 1974	17 2	1977 1976	18 2	1977 1976	59 10 2 6	1977 1976 1975 1 <del>9</del> 74	

purascens, Ostracoderma state of Peziza ostracoderma and Penicillium spp.

Curvularia protuberata Nelson and Hodges (Figs. 16, 17)

One specimen: on Festuca rubra (ES: Lappeenranta, 26. 9. 1976 S. V.). Isolated from Deschampsia and Phleum, in Canada and Scotland, respectively (cf. Ellis 1971).

Fusarium species occurred on about fourty grass throughout Finland (Table 4). The quantities of Fusarium species were generally small. Among the seven identified species, the most common were F. avenaceum, F. tricinctum, F. semitectum and F. poae. Species rarely encountered were F. graminearum Schwabe, found on five grass species; F. culmorum (W. G. Smith) Sacc., found on three grasses; and F. moniliforme found on Festuca pratensis (cf. Booth 1971).

Helminthosporium (Drechslera) species occurred on numerous grass species throughout Finland, but the quantities of the fungi were generally very small.

Helminthosporium biforme Mason and Hughes (Syn. Drechslera biseptata (Sacc. & Roum.), Richardson & Fraser) was found in about fifty seed lots including 32 grass species (Figs. 18, 20, 21). Most frequent among them were Anthoxanthum odoratum, Deschampsia flexuosa and Festuca rubra. Found to be very uncommon on Agrostis stoloniflera, and on Phleum pratense in one ley, and absent from seed of grasses produced in Finland (Mäkelä 1971, 1972 b). Reported on seeds of many grass species produced in different countries (Mäkelä 1972 b, Neergaard 1977), particularly on Dactylis glomerata (Tulloch and Leach 1972). The fungus is known to produce mycotoxins (Leach and Tulloch 1972).

Helminthosporium cyclops Drechsler (Syn. Drechslera verticillata Shoemaker, Bipolaris cyclops (Drechsl.) Sprague.) (Figs. 19, 26, 27). One specimen: on Sieglingia decumbens (U: Tvärminne 6. 8. 1975 A. P.). Found on seed of Poa pratensis in USA (Sprague 1950), and on seed of Bromus spp. (Cooke and Shaw 1952), as well as on different Graminae, e.g. Avena fatua, Bromus mollis and Poa pratensis (Shoemaker 1966). Conidia (74—) 100 (—138) X (16—) 18 (—22) μm, 6 to 10-septate.

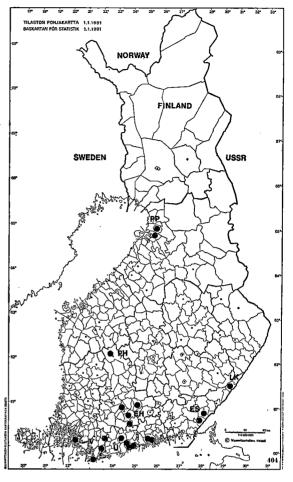


Fig. 18. The observations of *Drechslera biseptata* in grass seed.

Conidiophores (99—) 137 (—209) X 7—9  $\mu$ m, 3 to 7-septate (cf. Shoemaker 1966).

Drechslera dactylidis Shoemaker (Figs. 19, 22, 23).

Sixteen specimens found on eleven grass species throughout Finland: Agrostis vinealis (V), Dactylis glomerata (PS), Deschampsia caespitosa (EH), D. flexuosa (V, 2 exx., U, EK, EP), Festuca ovina (V, ES, 2 exx.), Nardus stricta (U), Phalaris arundinacea (EH), Phleum commutatum (Ks), Poa annua (V), P. nemoralis (V), and P. palustris (EH). Found on seed of Dactylis glomerata pro-

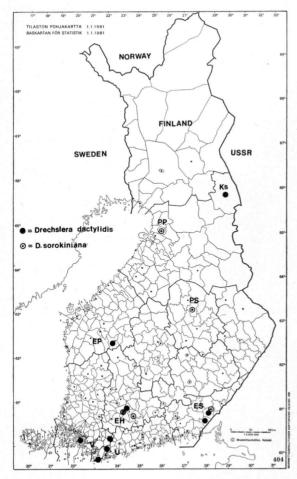


Fig. 19. The observations of *Drechslera dactylidis* and *D. sorokiniana* in grass seed.

duced in both Finland and Poland (Mäkelä 1971, 1972 b), as well as in eight other countries (Tulloch and Leach 1972). D. dacty-lidis resembles D. tetrarrhenae Paul (cf. Paul 1971).

Helminthosporium dictyoides Drechsler. Syn. Drechslera dictyoides (Drechsler) Shoemaker (Fig. 24).

Fifteen specimens found on nine grass species: Dactylis glomerata (PS), Deschampsia caespitosa (U, EP), Elymus arenarius (U), Festuca arundinacea (U, 2 exx.), F. ovina (U), F. pratensis (U), Holcus lanatus (U), Phleum

pratense (ES), Poa pratensis (U). Occurred in seed of Bromus inermis, Festuca pratensis, F. rubra and Lolium perenne produced in Finland (Mäkelä 1971). Common in leys throughout Finland (Mäkelä 1971). Cosmopolitan (Braverman and Graham 1960, Andersen 1974).

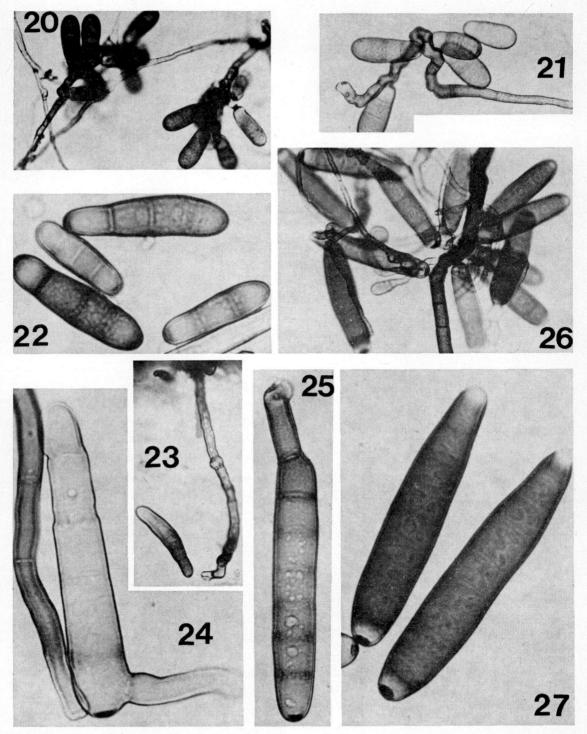
Helminthosporium sativum Pammel, King and Bakke. Syn. Bipolaris sorokiniana (Sacc. in Sorok.) Shoemaker, Drechslera sorokiniana (Sacc.) Subram. & Jain. Found on five grass species: Agropyron caninum (PP: Kiiminki), A. repens (EH: Janakkala), Deschampsia flexuosa (PS: Lapinlahti), Glyceria maxima (EH: Hattula), and Festuca rubra (ES: Lappeenranta).

Uncommon and infrequent on *Phleum* pratense and Agrostis tenuis in leys. Also found in seed of Bromus inermis, Festuca pratensis, Lolium perenne and Phleum pratense produced in Finland (Mäkelä 1971, 1972 b). Very common in low percentages in seeds of many Gramineae and largely seed-transmitted (cf. Neergaard 1977).

Helminthosporium siccans Drechsler. Syn. Drechslera siccans (Drechsler) Shoemaker (Fig. 25).

Found on four grass species: Agropyron caninum (U: Tvärminne), Deschampsia caespitosa (LK: Parikkala), Elymus arenarium (U: Porvoo rural district) and Festuca arundinacea (U: Helsinki). Found in seeds of Lolium perenne produced in Finland and fairly common on Lolium species in leys (Mäkelä 1971, 1972 a). Rather infrequent on seeds of different Gramineae (Chidambaram et al. 1973, Andersen 1974).

Helminthosporium vagans Drechsler. Syn. Drechslera vagans (Drechsler) Shoemaker. One specimen on Poa pratensis (U: Tvärminne, 26. 9. 1974 A. P.). The fungus was found to be moderately common on Poa pratensis in leys in Finland (Mäkelä 1972 a). Rather



Figs. 20—27. Drechslera species. 20, 21: D. biseptata on Alopecurus pratensis. 22, 23: D. dactylidis on Deschampsia flexuosa. 24: D. dictyoides f. sp. dictyoides on Melica nutans. 25: D. siccans on Deschampsia caespitosa. 26, 27: D. verticillata on Sieglingia decumbens.

Material: 20, 21: U, Riihimäki 4. 8. 1977 P.A. 22, 23: V, Suomusjärvi 25. 9. 1977 S.V. 24: EH, Hauho 6. 8. 8. 1977 P.A. 25: LK, Parikkala 10. 8. 1974 P.A. 26, 27: U, Tvärminne 6. 8. 1975 A.P. 20, 26: X 300. 21: X 500. 23: X 400. 22, 24, 25, 27: X 800.

common in seed of Poa pratensis and P. trivialis (Andersen 1974).

Coelomycetes, Melanconiales
Truncatella truncata (Lév.) Stey. (Fig. 28).
Found on six grass species: Alopecurus
aequalis (EH: Luhanka), Bromus hordeaceus
(U: Tammisaari), Holcus lanatus (U: Tvärminne), Melica nutans (EH: Hattula, 2 exx., U:
Porvoo rural district), Milium effusum (PS:
Lapinlahti), and Phleum pratense (EH: Hattula). Uncommon in leys of Phleum pratense
in northern Finland (Mäkelä 1981). The
fungus is one of the most pathogenic on red
clover seedlings in Finland (Ylimäki 1967) (cf.
Sutton 1980).

Sphaeropsidales

Camarosporium Schulzer sp. (Fig. 29).

One specimen on *Melica nutans* (U: Tvärminne, 29. 6. 1975 E. P.) (cf. Grove 1937, Sutton 1980).

Diplodia Fr. sp. (Figs. 30, 31).

One specimen on *Melica nutans* (U: Tvärminne, 29. 6. 1975 E. P.) (cf. Karsten 1890, Grove 1937, Neergaard 1977, Sutton 1980).

Hendersonia crastophila Sacc.

Fifteen specimens. Found on eleven grass

species (from U, EH, ES, EP, PS), moderately common. Very common on 33 grass species throughout Finland (Mäkelä 1977 a).

Phaeoseptoria festucae Sprag.

Moderately common on thirty grass species throughout Finland, particularly on *Calamagrostis*, *Festuca* and *Poa* species. The fungus is rare but moderately common on 12 grass species, being more common on cereals (Mäkelä 1977 a).

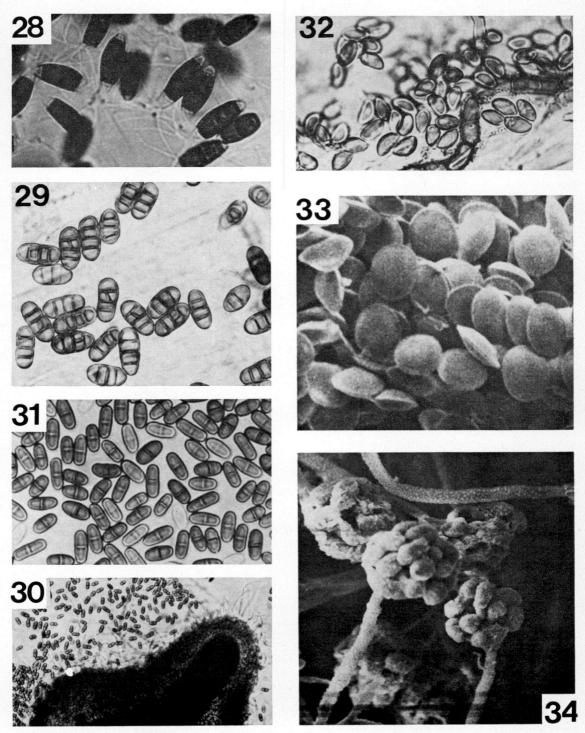
Phoma species were found to be very common on the seed of numerous grass species. Most common on Bromus hordeaceus (18 %) of seeds), B. inermis (6 %), Alopecurus pratensis (6 %), Milium effusum (5 %), Deschampsia caespitosa (2 %) and D. flexuosa (2 %). Found to be more common in the dry summers of 1975 and 1976 than in the rainy summers of 1974 and 1977. Phoma species found as parasites on the seeds of red clover (Salonen 1972), as well as on red clover seedlings (Ylimäki 1967). Phoma species are also sometimes reported on grasses (Sprague 1950, Mäkelä 1972 a) as well as on grass seed (Mäkelä 1972 b). Found on seed of Dactylis glomerata (Tulloch and Leach 1972).

#### CONCLUSION

This study gives a general view of the fungal microflora of wild grass seeds. In my opinion the fungi on the seeds of the wild grasses indicate the true Finnish microfungi better than those found on the seeds of cultivated grasses: much of the cultivated grass seed is imported. On the other hand, many genera of fungi contain as yet unidentified species in this study. It is known (Leach and Tulloch 1972) that many fungi on grass seed sporulate only in total darkness or after exposure to a diurnal cycle of near-ultraviolet radiation. Besides the number of seed investigated (100—200 seeds per lot) is rather little. However, many seed lots and grass

species were examined in this study, and each seed was also examined a few times with a stereomicroscope and a light microscope during the culture period of about one month.

The fungal material was found to be scanty. Most of the species of fungi were found to be common on many grass species but in very small quantities, and about twenty species of fungi were found to occur very rarely. Many species of microfungi were found only once. However, a few species of fungi, e.g. Alternaria tenuis and Cladosporium cladosporioides, were found to be very common.



Figs. 28—34. 28: Truncatella truncata on Bromus hordeaceus. 29: Camarosporium sp. on Melica nutans. 30, 31: Diplodia sp., on Melica nutans. 32: Coniella sp. on Alopecurus pratensis. 33: Arthrinium phaeospermum on Agrostis tenuis. 34: Stachybotrys atra on Agropyron repens.

Material: 28: U, Tammisaari rural district 5.7.1976 A.P. 29—31: U, Tvärminne 29.6.1975 E.P. 32: PP, Oulu 12.8.1976 M. & P.U. 33: U, Porvoo rural district 5.9.1976 A.P. 34: ES, Lappeenranta 28.8.1977 S.V. 33, 34: SEM, Viikki. 28, 31: X 850. 29, 34: X 1000. 30: X 200. 32: X 750. 33: X 1300.

Many of the rare fungi were found, e.g. Scopinella solani, Ryparobius polysporus and Drechslera verticillata. The reason for this might be that many grasses carry the specific microflora or at least some specific species of the fungus. As wild grasses are common throughout Finland (Hultén 1971), this makes the spread of grass disease possible. Furthermore many species of wild grass grow together. It is generally known that a monoculture is more susceptible to disease than a mixed culture.

In addition to the weather conditions, the combination of grass species and type of culture are an important factor affecting the occurrence of the mycoflora of grass seed.

The species of fungi were therefore found to be more numerous in the very rainy summer of 1977 than in the dry summer of 1975 in this study as well as those of, e.g. Salonen (1972), and Tulloch and Leach (1972).

It is very difficult to make an absolute distinction between parasites and saprophytes. In fact, one's knowledge of many fungi is so superficial that one cannot with certainty omit any species as being of no importance (cf. Tulloch and Leach 1972).

Many fungi species are cosmopolitan. This concept is also confirmed by the results of this study (cf. Mäkelä 1972 b, Tulloch and Leach 1972, Neergaard 1977).

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#### SELOSTUS

# Luonnonvaraisten heinien siementen sienistä

## KAIHO MÄKELÄ

# Maatalouden tutkimuskeskus

Tutkimus perustuu vuosina 1974—1977 Helsingin yliopiston kasvitieteellisen puutarhan toimesta kerättyyn siemenaineistoon, joka käsitti 63 luon-

nonvaraista heinälajia, yhteensä 284 siemenerää. Näytteitä oli kautta maan, valtaosa niistä Uudeltamaalta, Etelä-Hämeestä ja Etelä-Savosta. Siemenet, 100 kpl erää kohti, kasvatettiin Jacobsenin idätysaltaassa. Niistä laskettiin itävyys sekä tutkittiin sienilajisto mikroskooppisesti ja mikrovalokuvausta käyttäen.

Sieniä todettiin yli 90 lajia, ja ne kuuluivat 60 sukuun. Valtaosa, n. 60 lajia, kuului luokkaan Deuteromycotina, Vaillinaissienet. Luokkaan Ascomycotina, Kotelosienet, kuului 25 lajia.

Sienten lukumäärä vaihteli suuresti eri heinälajeissa ja samankin heinän näyte-erissä. Runsas sienilajisto oli mm. nuokkuhelmikän (Melica nutans) siemenissä, yhteensä 46 lajia (vaihtelu eri erissä 7—22) ja juolavehnän (Agropyron repens) siemenissä yhteensä 37 sienilajia (vaihtelu 14—24). Sen sijaan niittynurmikan (Poa pratensis) siemenistä löytyi yhteensä vain 15 sienilajia (vaihtelu erien välillä 5—12).

Sienilajien lukumäärä ja sienten kokonaismäärä vastasivat useimmiten toisiaan. Toisaalta yksittäisten sienten määrät tutkituissa siemenissä olivat vähäisiä, muutamia sienilajeja lukuun ottamatta.

Myös vuosien väliset eroavuudet sienten määrässä olivat suuria. Viileänä ja sateisena kesänä 1977 oli sienilajeja keskimäärin noin kaksi kertaa enemmän ja sienten kokonaismäärä noin 40 % suurempi kuin lämpimänä ja kuivana kesänä 1975. Monet haitallisiksi todetut sienet, kuten Claviceps purpurea, Fusarium- ja Helminthosporiumlajit samoin kuin eräät kotelosienet, mm. Pleospora- ja Leptosphaeria- lajit esiintyivät runsaimmin juuri kosteina kesinä 1974 ja 1977.

Seitsemästä todetusta *Helminthosporium*-lajista oli yleisin, 32 heinälajilla esiintyvä *H. biforme.* Fusarium-lajeja todettiin myös seitsemän ja niistä olivat yleisimpiä *F. avenaceum, F. tricinctum, F. semitectum, F. poae* ja *F. gramineurum.* 

Eräitä harvinaisuuksia, kuten Scopinella solani, Curvularia protuberata ja Helminthosporium cyclops, esiintyi muutamissa siemenerissä. Valtaosa sienilajeista oli kosmopoliitteja.

Siementen itävyys vaihteli suuresti heinälajeittain ja vuosittain. Hyvin itivät mm. timotein (Phleum pratense); mäkikattaran (Bromus hordeaceus) ja nurmikkajien (Poa) siemenet. Sen sijaan mm. siniheinän (Molinia caerulea), kastikka-lajien (Calamagrostis) ja idänkattaran (Bromus inermis) siemenet itivät erittäin heikosti. Lämpimän ja kuivan kesän 1975 siementavara iti parhaiten, viileän ja märän kesän 1977 huonoiten.

# Seria PHYTOPATHOLOGIA N. 80 — Sarja KASVITAUDIT n:o 80

# FUSARIUMS OF THE POTATO IN FINLAND I. ON THE FUSARIUM SPECIES CAUSING DRY ROT IN POTATOES

#### ESKO SEPPÄNEN

Seppänen, E. 1981. Studies on Fusariums of the potato in Finland I. On the Fusarium species causing dry rot in potatoes. Ann. Agric. Fenn. 20:156—160. (Agric. Res. Centre, Inst. Pl. Path. SF-01300 Vantaa 30, Finland).

Fourteen Fusarium species were isolated and identified from potato tubers with dry rot symptoms. In preliminary pathogenicity tests all these species proved to be pathogenic to cultivar Bintje. In this material Fusarium avenaceum and F. solani var. coeruleum were predominant, followed by F. culmorum and F. sulphureum.

Index words: Potato, dry rot, pathogen, Fusarium.

## INTRODUCTION.

Fusarium species have been known as the cause of potato tuber rots since the last century, but their importance has increased simultaneously with the mechanization of potato production, because of their character as wound parasites. In Finland this problem increased from 1975 onwards, when large amounts of seed potatoes were imported from a number of western European countries. The present paper deals with studies on the Fusarium species pathogenic to potatoes isolated from the imported and native stocks.

Although there is ample published information about potato tuber rots and about the pathogens causing them, the appropriate starting point for a short review is the monograph of the genus Fusarium by Wollenweber and Reinking (1935). It gives a good general idea of the diseases and of the Fusarium species as causal organisms. Wollenweber and Reinking presented four com-

mon pathogens: Fusarium trichothecioides Wollenw., F. coeruleum Sacc. (according to Booth 1971 F. solani var. coerulem (Sacc.) Booth), F. sambucinum Fuckel f. 6 (F. sulphureum Schlecht.), and F. avenaceum (Corda ex Fr.) Sacc., which were still the dominant species in the 1970s (cf. Boyd 1972). F. solani (Mart.) App. et Wr. var. striatum (Sherb.) Wr. (F. solani) and F. orthoceras App. et Wr. var. longius (Sherb.) Wr. (F. oxysporum) were also mentioned as pathogens of potato tubers.

According to Booth (1971), Fusarium sambucinum Fuckel was apparently presented by Sherbakoff (1915) under the name F. discolor. McKee (1952) regarded it as non-pathogenic, but later a number of authors (Benken et al. 1968, Gorodetskii 1970, Stubbs 1971, Kutova 1976, Seppänen 1980) considered it pathogenic. It is relatively often reported from Eastern Europe.

Jamalainen (1943 a, 1943 b, 1944) isolated

eight species from potato tubers. Besides those named above (F. avenaceum, F. sambucinum, F. solani and F. solani var. coeruleum), he identified the species F. culmorum (W. G. Smith) Sacc., F. arthrosporioides Sherb., F. merismoides Corda and F. scirpi var. acuminatum (according to Booth 1971 F. acuminatum). He did not regard all of them as primary pathogens, but each of them has later been proved to be pathogenic. F. culmorum, which McKee (1952) did not consider a pathogen, was reported as pathogenic by Janke (1976), Kutova (1976), Götz and Pett (1977), Seppänen (1980) and Tivoli and Jouan (1980). Tivoli and Jouan described it under the name F. roseum var. culmorum. F. arthrosporioides, which was proved to be pathogenic by McKee (1952), closely resembles F. avenaceum and is often confused with it. F. merismoides was proved to be pathogenic by Førsund (1980), and F. scirpi var. acuminatum (F. acuminatum) was proved to be a weak pathogen of cv. Bintje by Seppänen (1980).

A number of other species have also been reported as pathogens of the potato tuber: F. tricinctum (Corda) Sacc. (McKee 1952, Jan-

ke 1976, Seppänen 1980); F. sporotrichioides Sherb. (Uppstone 1970, Abdel-Moniem 1977, Seppänen 1980); F. flocciferum Corda (Booth 1971); F. redolens (Wollenw.) Gordon (F. oxysporum var. redolens) (Stubbs 1971, Seppänen 1980); F. sambucinum Fuck. var. minus Wr. (Dorozhkin and Mikhal'chyk 1975), synonymous with F. sambucinum var. coeruleum according to Booth; F. semitectum Berk. & Rav. (Janke 1976); F. sarcochroum (Desm.) Sacc. (Kutova 1976), synonymous with F. sambucinum according to Booth (1971); F. concolor Reinking (Abdel-Moniem 1977); F. equiseti (Corda) Sacc. (Rai 1979); and F. graminearum Schwabe (Seppänen 1980, Tivoli and Jouan 1980) for which Tivoli and Jouan used the name F. roseum var. graminearum.

According to Booths (1971) system of classification 20 species, excluding synonyms, are reported as pathogens of the potato tuber. Some confusion in identification is possible, because of the great variation in certain species; i.e. some fungi may be known by the same name and a single fungus may have several names.

## MATERIAL AND METHODS

# Material

In connection with the seed potato imports of 1975, a total of 73 tuber samples were analysed to investigate the occurrence of potato gangrene and Fusarium dry rots. During subsequent years, about 300 tuber samples were collected from different parts of the country, partly from stocks originating from the imported stocks and partly from farms having no contact with the imported stocks. No schedule was used in the collection of the samples, and some were received from farmers who sent tuber samples demonstra-

ting a »new disease». In addition to these, 75 samples of 300 tubers each from northern Finland were analyzed in 1980 to investigate the importance of different fungi causing storage diseases.

#### Isolation and identification

The pathogens were isolated using conventional methods. Tubers with dry rot lesions were cut into pieces, disinfected and placed in Petri dishes on moist filter paper at room temperature. Within a few days the fungi

had grown sufficiently for isolation. The isolates were grown on potato dextrose agar (PDA) or potato sucrose agar (PSA), and only the latter was used for identification. The PDA was a Difco product and the PSA was prepared according to Booth (1971). Each isolate identified was raised from a single hypha of the fungus. For identification, the fungi were grown on PSA at a temperature of 22—25°C, 35—40 cm below fluorescent tubes with a 12 hour light and dark rhythm. The identification is based on Booths (1971) system. For determination of optimum

growth rate, the fungi were grown on PSA in constant darkness.

When identifying fungi such as the genus Fusarium, where the variation within a certain species is large and the differences between the species small, some difficulties may arise. Characters such as growth rate (GR), growth appearance, and even the colour of colonies varied to some extent and they must be considered as fairly approximate. In this study the main emphasis in identification was on the production, size and shape of spores.

## RESULTS AND DISCUSSION

As the material of this study was rather large and originated from a number of countries, 14 Fusarium species pathogenic to potatoes were found. They are presented in Table 1 with their relative frequencies and some of their characters.

Most of the fungi were identified easily according to Booths (1971) classification. The only real problems which caused difficulties in identification were the single isolates of Fusarium sambucinum Fuckel var. coeruleum and F. trichothecioides. The former, which

Table 1. Fusarium species isolated, identified and proved to be pathogens of potato tubers, their approximate frequency in the material, growth rate on PSA at 22°C, approximate optimum and maximum growth temperatures on PSA, and estimated pathogenicity in cv. Bintje.

Species	Frequency	Growth rate	Optimum temp.	Max. temp.	Pathogen- icity
Section Martiella					
Fusarium solani (Mart.) Sacc. emend Snyder & Hanser	1 < 1	30	28	36	weak
F. solani var. coeruleum (Sacc.) Booth comb. nov.	30	25	25	30	strong
Section Sporotrichiella					
F. tricinctum (Corda) Sacc.	< 1	40	25	32	weak
Section Arthrosporiella					
F. sporotrichioides Sherb.	< 1	42	28	34	moderate
F. avenaceum (Corda ex Fr.) Sacc.	30	52	2.5	32	strong
Section Elegans					
F. oxysporum Schlecht. emend Snyder & Hansen	5	44	28	34	weak
F. oxysporum var. redolens (Wollenw.) Gordon	< 1	38	30	34	weak
Section Gibbosum					
F. acuminatum Ellis & Everhart	< 1	30	24	32	weak
Section Discolor					
F. culmorum (W. G. Smith) Sacc.	15	80	25	34	moderate
F. graminearum Schwabe	< 1	90	25	34	strong
F. sambucinum Fuckel	5	42	27	33	moderate
F. sambucinum Fuckel var. coeruleum Wollenw.	< 1	58	25	32	strong
F. sulphureum Schlecht.	10	62	25	34	strong
F. trichothecioides Wollenw. in Jamieson & Wollenw.	< 1	62	25	34	strong

had spore production and size and shape of conidia as presented by Booth, deviated in the colour of the colony. This was greyish at the beginning of growth but rapidly turned into peach-beige and greyish-light brown after 10 days as a consequence of high chlamydospore production. The colony of the latter was peach to salmon when young (very similar to F. sulphureum) but later became greyish-light brown. At about 20 days a number of dark brown, dot-like sporodochia scattered around the dish developed, as stated by Booth. The growth rate was far slower than that reported by Booth. The conidia also deviated in being longer and narrower, as described by Wollenweber and Reinking (1935). The symptoms it caused in cv. Bintje and its pathogenicity were very similar to those of F. sulphureum. Much attention was

paid to finding an isolate of *F. arthrosporioides*, which has been found earlier in Finland, but there was none present.

The total number of Fusarium spp. identified includes nearly all of the species reported as pathogens of potatoes, and includes most of the strong pathogens. In the light of this study, it seems that F. avenaceum, F. solani var. coeruleum, F. culmorum and F. sulphureum are the most common species causing dry rots in Finland. The results of studies carried out on their pathogenicity under different environmental conditions and in different cultivars are presented separately (Seppänen 1981 a, b).

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## **SELOSTUS**

# Perunan kuivamätää aiheuttavista Fusarium-lajeista Suomessa

#### ESKO SEPPÄNEN

### Maatalouden tutkimuskeskus

Tutkimuksessa on selvitetty kuivamätää aiheuttavaa Fusarium-lajistoa. Aineistona oli osaksi 1975 maahantuodut siemenperuna-erät, osaksi niistä lisätyt tai täysin kotimaiset perunakannat. Sienten eristäminen tapahtui tavanmukaisin keinoin. Lajien määrittäminen on tehty Boothin (1971) luokituksen mukaan.

Aineistosta määritettiin kaikkiaan 14 perunan mukuloissa patogeenista lajia: ne, samoin kuin niiden suhteellinen yleisyys, kasvunopeus perunasakkaroosiagarilla, kasvun optimi- ja maksimilämpötilat sekä patogeenisuus Bintjessä on esitetty taulukossa 1. Tämän tutkimuksen mukaan merkittävimmät varastotauteja aiheuttavat Fusarium-lajit ovat F. avenaceum ja F. solani var. coeruleum, myös F. sulphureum ja F. culmorum ovat kohtalaisen yleisiä ja niilläkin lienee käytännön merkitystä. Muut lajit ovat verraten harvinaisia.

# Seria PHYTOPATHOLOGIA N. 81 — Sarja KASVITAUDIT n:o 81

# FUSARIUMS OF THE POTATO IN FINLAND II. ON THE GROWTH OPTIMA OF FUSARIUM SPECIES IN TUBERS OF CV. BINTJE

#### ESKO SEPPÄNEN

Seppänen, E. 1981. Fusariums of the potato in Finland II. On the growth optima of Fusarium species in tubers of cv. Bintje. Ann. Agric. Fenn. 20: 161—176. (Agric. Res. Centre, Inst. Pl. Path., SF-01300 Vantaa 30, Finland.)

The growth of fourteen Fusarium spp. at temperatures of 6, 12, 18, 24, 30 and 36°C and under different conditions of relative humidity (RH) was studied using artificial infection of Bintje tubers. The optimum growth conditions of each Fusarium studied were as follows: Fusarium sambucinum Fuckel var. coeruleum Wollenw. favoured low temp. (10-12°C) and low RH (40-50 %), F. sulphureum Schlecht. and F. trichothecioides Wollenw, in Jamieson and Wollenw, also favoured low RH, but had two optimum temperatures (10-12°C and 24-27°C), whereas F. culmorum (W. G. Smith) Sacc. and F. sporotrichioides Sherb. demonstrated nearly the same two temperature optima but favoured high RH (90-100 %); F. graminearum Schwabe and F. solani var. coeruleum Sacc. Booth comb. nov. grew best at a rather high temperature (about 20°C) and high RH (90-100 %); F. avenaceum (Corda ex Fr.) Sacc. and F. tricinctum (Corda) Sacc. favoured high temperature (nearby 30°C) and high RH; F. solani (Mart.) Sacc. emend Snyder & Hansen and F. oxysporum var. redolens (Wollenw.) Gordon were only studied in preliminary tests and seemed to favour high temperature and high RH. The isolates of F. acuminatum Ellis & Everhart and F. sambucinum Fuckel used in preliminary tests were not sufficiently pathogenic to indicate any particularly favourable conditions. Only some fungi demonstrated nearly the same optima when cultivated in Bintje tubers or on potato sucrose agar (PSA). That certain species have two optimum temperatures is presumably because the resistance reactions displayed by tubers are at an optimum between 15 and 25°C.

Index words: Potato, cv. Bintje, growth of Fusarium spp., optimum temperature, relative humidity, dry rot.

# INTRODUCTION

Thermotherapy is a well-known method of controlling potato gangrene during a wound healing period. Knowledge of its applica-

tion in the control of potato dry rots is superficial; this is perhaps due to the relatively limited information available about the reactions of Fusarium species under different environmental conditions. The aim of this study was to clarify whether it might be possible to develop a method to control Fusarium induced rots. Some of the results of this study have already been presented (Seppänen 1980 a, b) but are not referred to in the following literature review.

The first studies on the importance of environmental conditions were carried out at the beginning of this century. To date, however, the information is superficial and is limited to the general growth trends of some strong pathogens. Four Fusaria dominate as pathogens of potato tubers.

Fusarium solani var. coeruleum (F. coeruleum) is found throughout the world and has been the object of greatest interest. Wollenweber and Reinking (1935) reported it as growing at between 3 and 30°C, the most favourable temperature being 15—28°C, especially at relative humidities (RH) of 50—80 %. Later, Moore (1945), McKee (1954) and Tivoli and Jouan (1980) ascertained the optimum growth temperature to be 15—20°C. In trials carried out by Weiss et al. (1928), high RH (90—98 %) proved to be more favourable than lower RH (70—80 %), and Moore (1945)

also considered high moist conditions favourable

Fusarium avenaceum favours higher temperatures, 20—25°C (McKee 1954). There is no information about its demand for humidity as a pathogen of the potato.

Fusarium trichothecioides is able to infect potato tubers at a range of temperatures (2—30°C) and grows well at temperatures of 5—25°C (Sanford 1924, Ref. Wollenweber and Reinking). Weld and Wollenweber (1912) established that it penetrates potato tubers more rapidly under dry than under moist conditions in trials carried out at 10—12°C. The same result was found in trials by Weiss et al. (1928), who stated that spore infection, in particular, favoured drier conditions than mycelia plug infection. According to Goss (1921) it favours high RH.

Fusarium sulphureum (F. sambucinum Fuckel f. 6) is often reported as a pathogen of potato tubers in North America and Central Europe. However, there is hardly any information about its needs concerning temperature and humidity. The same situation applies to the other species reported as being more or less weak or occasional pathogens.

## MATERIAL AND METHODS

# Material

Since 1975, we have isolated fourteen Fusarium species pathogenic to potato tubers. They were identified according to Booth's classification (Seppänen 1981 a). One or more isolates were taken from each species for this study.

For the tests, each isolate was grown in Bintje tubers to maintain its pathogenicity. Each isolate used was raised from a single hypha of the fungus. The fungi were grown on potato sucrose agar (PSA) prepared according to Booth (1971), first at room temperature and after some days at a temperature of about 6°C. The fungi were used in their period of profuse spore production, usually 2—5 weeks after culturing.

The tests were carried out with tubers of cv. Bintje, which is sufficiently susceptible to indicate the reactions of the fungi under different environmental conditions. The tubers used were grown according to common practice, the stock of each season being of rather even quality.

#### Methods

The growth of the Fusaria or their separate isolates under different environmental conditions was studied using an adaption of the method of Langton (1971). A wound 5 mm in diameter and 2 mm deep was made with a cork borer at the mid-point between the heel and rose end of washed and dried tubers of the same size class. A mixture of pure cultures of the fungi and the remaining agar medium was used as inoculum. The age of the colonies varied from two to five weeks depending on the spore production of each The wounds were filled with fungus. inoculum and left uncovered. The method was equally as effective as Langton's plug method.

The number of tubers used for each treatment was 10 with three replicates. Tuber infection was scored after 20 days incubation. The tubers were halved longitudinally through the infection locus, and the growth of the fungus at the cut tuber surface, i.e. the radial growth in the peel (and beneath it if greater) and the deepest axial growth were measured. In general, only the average figures for radial and axial growth or their means are presented.

The infected tubers were incubated in plastic boxes of 10 I which were left uncovered if the RH of the incubating chamber was suitable. We had great difficulties in regulating the humidity at different temperatures and so we used only two or three values of RH. The high humidity, RH 95  $\pm$  5% was achieved by putting water and an extra bottom into the box and enclosing the whole experiment in a plastic bag. According to our experience the lower amount of oxygen

in the bags than in the chamber had no essential influence on the results, because the incubation time was only 20 days. The temperatures used were 6, 12, 18, 24, 30 and 36°C. The approximate optimum growth temperatures of the fungi on PSA were determined for comparison with those obtained in potato tubers. The fungi were grown on PSA medium in the dark.

In many tests it was impossible to use all these temperatures simultaneously, and, for drawing graphs, the result were transformed on the basis of the results of one temperature with the high RH, which was common to the all treatments.

All the results were analyzed using variance analysis and the LSD values were calculated with Tukey-Hartley tables.

The tests were carried out over several years and throughout the storage season, resulting in a certain variation in the tuber material. In some cases in which the optimum is between two temperature values used, the variation is extremely striking. Throughout the work we have used only one potato cultivar and as a rule a single isolate of each fungus. It is possible that a different cultivar and other isolates might produce results which deviate from those presented here. The importance of a particular isolate within a species is probably not great, because we obtained almost the same results when the number of isolates was increased. On the other hand, the test cultivar might be of major importance because different cultivars react somewhat differently at different temperatures. In my experience the basic requirements for this kind of study are a very susceptible cultivar and a virulent fungus isolate.

The fundamental problems of this investigation can be considered solved. The work will, however, continue in order to clarify some details and the possible importance of the cultivar.

# RESULTS AND DISCUSSION

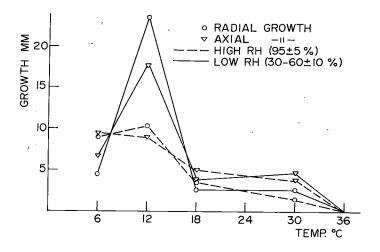
The results are presented in Tables 1—12 and Figures 1—8, starting from fungi demonstrating low optimum values. For critical examination of some figures it must be remembered that the curves representing the growth at lower RH are not based on results obtained under absolutely controlled conditions of RH. As a rule, the degree of infection was 100 %, except in the treatments at 36°C that were excluded from the variance analysis.

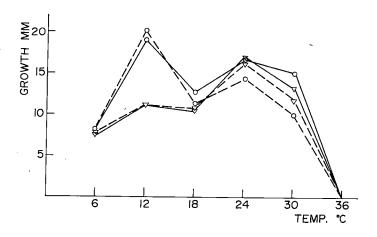
Fusarium sambucinum Fuckel var. coeruleum Wollenw. The fungus is pathogenic within wide temperature (about 5 to 30°C) and RH ranges. However, it was a strong pathogen within quite narrow temperature limits, the optimum being nearby 12°C, and at temperatures of 18°C or higher it was incapable of progressive advance (Table 1. Fig. 1). This fungus clearly grew better in the lower values of RH studied than in the highest one (Table 3). At temperatures of 6 and 12°C the symptoms produced by this and by isolates of F. sulphureum were fairly similar.

Fusarium sulphureum Schlecht. Two isolates of this fungus were used, one (7523-8) able to produce a red pigment when growing in the dark on PDA (potato dextrose agar, Difco) and the second (7592-4) incapable of this. In other factors, such as the growth on PSA, microscopical identification, symptoms produced in Bintje and in their reactions to different growth conditions, they behaved fairly similarly. This fungus is strong pathogen over wide temperature and RH ranges. A surprising result was that it has two temperature optima, the lower at about 10-12°C and the higher between 24 and  $30^{\circ}$ C (Table 2 and Figures 1 and 3). In Bintje tubers, the radial growth was dominant at the lower optimum and the axial growth was more rapid at the higher. The two optima are perhaps explained by variations in the resistance reactions of the tuber according to temperature. The result was confirmed with a number of isolates incubated in an RH of 95  $\pm$  5 % (Fig. 3), although the higher optimum particularly appeared at low RH. This fungus is a strong pathogen in both high

Table 1. The growth of  $Fusarium\ sambucinum\ var.\ coeruleum\ (isolate\ 7544—5)\ under\ different\ environmental\ conditions.$  Incubated for 20 days.

Temp. °C	RН	March	1979	April	1979	March	1980
	% 	Radial	Axial	Radial	Axial	Radial	Axial
6	50 ± 10	4,5	6,5		. <u>—</u>		
6	$95 \pm 5$	9,0	9,3		_	3,3	5,6
12	$40 \pm 10$	23,6	17,8	20,7	14,9		
.2	$70 \pm 10$	18,5	16,6	20,2	17,9		
2	$95 \pm 5$	10,3	8,9		<del> , . ,</del>	7,8	8,5
8	$30 \pm 10$	3,0	4,7	2,7	4,5		5,0
8.	50 ± 10	2,5	3,7	2,7	4,6	_	
8	$95\pm5$	3,6	5,1	<del></del>		2,3	3,6
4	95 ± 5			_		2,0	3,6
0	$70 \pm 10$	2,6	4,7	_		2,0	5,0
0	95 ± 5	2,6	4,1		-	1,6	3,9
	F	454,38 ***	161,67 ***	983,06 ***	136,91 ***	39,87 ***	30,45 ***
	PMI	£ 1,8	1,2	1,1	1,9	1,3	1,5





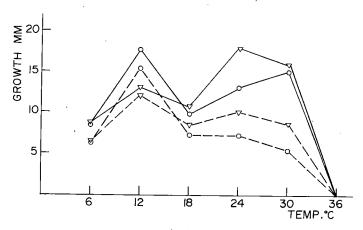


Fig. 1. The growth of Fusarium sambucinum var. coeruleum (upper), F. sulphureum (middle) and F. trichothecioides (lower) under different environmental conditions.

Table 2. The growth rate (mm) of two isolates of Fusarium sulphureum under different environmental conditions, the trials were carried out in 1979. Incubated for 20 days.

Radial Axial Radial Radial Axial Radial Radial Radial Axial Radial Radia Radial Radia R		January		March	e	April		May		October	H
Coctobe   Coct	I H	'	Axial	Radial	Axial	Radial	Axial	Radial	Axial	Radial	Axial
16.5    11.5    15.6    9.8   10.3	8										
0 4,7   5,6   4,8   11,0     24,2   21,9	<b>.</b>	-	-		-			1	I	8.1	7.2
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		4,7	5,6	ج موري موري	10,0	1			ļ	, e	6.7
16,3    11,5    15,3    15,2    24,2    21,3		2,1	2,5	14,8	1,1,0	٦ [	1 -	İ		26	.
1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1,		1	i	25,3	17,2	7.4.7	ST, 9   - 6.	Ì	ļ	i d	- 00
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		16,3	11,5	19,9	15,2	23,0	19,5	i	1	2,6	-
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		7.0	5,2	16,0	11,7		1	-	Ī	14,0	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		6.2	5,5	15,6	12,5	10,2	14,1	1 ;	-   ;	= }	<u>-</u> כן
1, 2, 3, 4, 6   9, 1   9, 6		-	- }	12,7	13,6	10,3	13,4	10,0	7,9	5,	2,5
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	- L	4 6		9.6	1	1	16,8	12,6	6,4	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	-1-		261	- (5	<u>-</u>	]	I	14,2	18,1	13,3	19,6
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	H.	ļ	1				1	1.7.0	17.3	0.5	7.2
F   320,95 *** 53,96 ***   19,3   16,7	+1	I	i	ŀ	1 }	ļ				17.7	21,2
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	+1	i	]	19,3	16,7	1	I		1,4,1	-6,4	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		ı	l	7,0	8,5	1		14,4	F.63	7,1	0,0
January         June         Octobe           10         9,0     9,0     15,2   8,8   15,3   15,9   15,9   15,9   15,9   15,9   15,3	F		53,96 *** 1,2	77,83 ***	41,82 ***	178,50 ***	46,94 *** 2,0	35,41 *** 1,6	41,21 *** 1,9	40,96 *** 2,0	78,30 *** 1,8
± 10         9,0             9,0             5,0   6,8           6,8           8,5           7,6           7,6           7,6           7,6           7,6           7,6           7,6           7,6           7,6           15,9           8,5           15,9           8,5           15,0           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7           15,7	Isolate 7592—4									(	į
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$		January	<b>A</b>			June				Octob	er
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	+	0.0	9.0			5,0	6,8			3,51	7,1
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	۱+	3.4	6.4			7,7	7,4			96)	6,0
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	- -				21,6	16,7			15,9	13,9
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	16,1	0,6			17,8	14,4			15,1	8,77
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	7.0	9,4			8,5	8,2			12,9	12,5
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	, K	7.2			8,6	7,6			1,7,	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	10.3	13,9			16,5	20,5			171 18, 18 19	18,0
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	1+	7.8	10.6			7,8	11,5			ر مورو م	4,0
F 71,38 *** 14,51 *** 67,63 *** 40,34 *** 50,06 *** 1,7 2,1 2,2 2,1 2,2	1+	- }	-			15,0	15,0			16,0	21,0
F 71,38 *** 14,51 *** 50,06 *** 50,06 *** 1,5 2,0 1,7 2,2 1,7	1+1 a ro	I	1			5,9	10,5	!		1	1
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		***************************************	***			67.63 ***	1			50,06 ***	54,10 ***
	TSD TSD	1.5	2,0			2,1				1,7	2,1

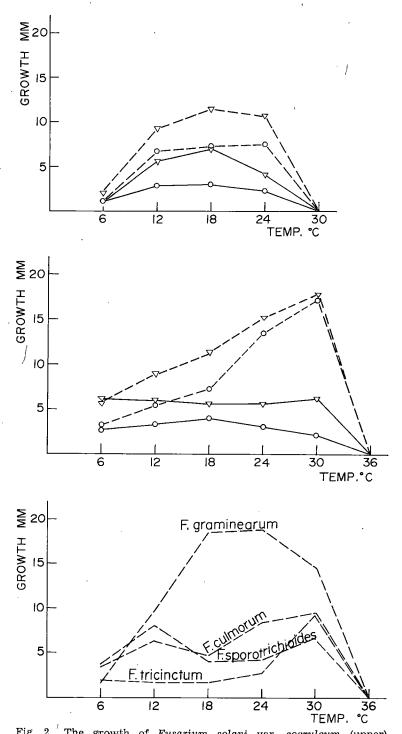


Fig. 2. The growth of Fusarium solani var. coeruleum (upper), F. avenaceum (middle) and F. graminearum, F. culmorum, F. sporotrichioides and F. tricinctum (lower) under different environmental conditions. The lower curves represent averages of radial and axial growth at high RH.

Table 3. The influence of the RH in the incubation chamber on the growth of two isolates of Fusarium sulphureum and on F. sambucinum var. coeruleum at a temperature of 12°C. Incubated for 20 days.

•		Fusarium su	lphureum		F. sambucinum v	. coeruleum
	7523—8	7592-	-4		7544—	5
RH %	Radial	Axial	Radial	Axial	Radial	Axial
40 ± 10	25,3	17,2	25,4]	20,9]	23,6	17,8
$70 \pm 10$	19,9	15,2	19,1	17,3	18,5	16,6
95 ± 5	16,0	11,7	15,9	11,4	10,3	8,9
F	27,08 ***	62,30 ***	113,34 ***	36,16 ***	158,43 ***	90,73 ***
$LSD_{5\%}$	3,1	1,2	1,6	2,7	1,8	1,8

Table 4. The growth of Fusarium trichothecioides (76277—4) under different environmental conditions. Incubated for 20 days.

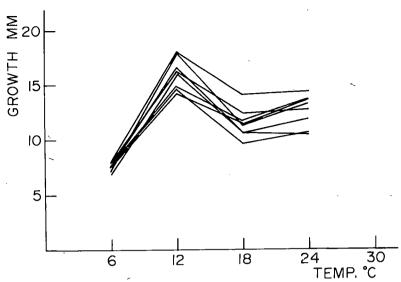
Temp.	RH	February 1	1979	June 19	79	May 19	980
°C	.% —	Radial	Axial	Radial	Axial	Radial	Axial
6	60 ± 10	13,4	8,6	4,4	5,7	7,0	8,4
6	95 ± 5	11,6	8,8	5,1	5,9	7,9	8,3
12	$55 \pm 10$	17,8	12,9		<del>-</del>	<b>—</b> .	— .
12	$75 \pm 10$		_ `	20,5	12,5	22,2	18,5
12	95 ± 5	19,1	10,7	20,9	11,2	21,0	11,8
18	$50 \pm 10$	16,2	13,1	7,9	8,0	13,9	17,6
18	$95 \pm 5$	11,6	8,2	8,9	7,7	13,9	16,6
24	$50 \pm 10$	20,1	15,7	8,6	11,7	21,3	23,5
24	95 ± 5	12,8	15,3	14,3	14,8	16,3	18,4
30	$50 \pm 10$	—,·	<del></del>	18,7	16,5	8,4	11,1
30	95 ± 5		_	13,1	13,1	8,3	11,2
	F LSD	46,22 *** 0,7	39,75 *** 0,8	80,10 *** 0,7	58,18 *** 0,9	77,04 *** 0,9	49,95 *** 0,9

and low RH, but clearly favoured dry conditions (Table 3). Under favourable environmental conditions, the fungus grew more than one millimetre a day in the tuber. The symptoms which developed under low temperature conditions (6 and 12°C) were quite different from those developed at high temperatures (24 and 30°C). At the former temperatures the cut surface was usually greyish brown, moist and firm, and almost no cavity was formed within 20 days, whereas typical powdery white rot, with a dark barrier between rotten and healthy flesh, developed under the latter conditions (Fig. 7).

Fusarium trichothecioides Wollenw. in Jamieson and Wollenw. behaved like F. sulphureum isolates; the optimum growth

conditions as well as the symptoms produced in Bintje were fairly similar to those produced by *F. sulphureum* (Table 4 and Fig. 1). The optimum temperatures were 12°C and between 24 and 27°C. Its requirement of low RH was not as clear as with *F. sulphureum*, and further tests are needed for more exact determination. Hornok (1980) has recently established a close chemical relationship between these two species.

Fusarium sporotrichioides Sherb. has two optimum temperatures, too. The lower is at about 12°C and the higher at about 30°C (Table 5 and Figures 2 and 6). Its RH requirement in the light of these trials seems to be about 70—80 %, but further trials are needed to obtain reliable information. The



TEMP. Fig. 3. The growth (average of radial and axial) of 8 Fusarium sulphureum isolates at different temperatures and in 95  $\pm$  5 % RH.

Table 5. The growth of Fusarium sporotrichioides under different environmental conditions. Incubated for 20 days.

	DIT	March	1980	April 1	1980
remp. C	. RH %	Radial	Axial	Radial	Axial
 6	95 ± 5	2,2	4,1	3,6	4,9
	$75 \pm 10$	<del>_</del> '	_ `	11,3	11,3
2	$95 \pm 5$	9,1	7,2	8,2	7,1
3	95 ± 5	3,9	4,4	3,8	3,8
1	$95 \pm 5$	3,2	4,6	4,0	4,8
ĵ	$50 \pm 10$	-,-		3,2	3,3
)	95 ± 5	6,7	9,6	3,9	6,8
	F	14,91 ***	12,40 ***	74,08 ***	45,69 ***
	LSD	2,3	2,1	1,1	1,2

Table 6. The growth of Fusarium culmorum under different environmental conditions. Incubated for 20 days.

	o. RH %	March 1980		April 1980	
Tem] °C		Radial	Axial	Radial	Axial
6	95 ± 5	1,9	4,6	3,1	5,0
12	$70 \pm 10$	<del>-</del> , ,		4,4	5,4
2	95 ± 5 95 ± 5	5,4   3,9	7,6   5,3	5,0 3,8	6,8   5,8
.8 24	45 ± 10		— ·	1,3	1,7
24	95 ± 5	8,3	12,9	4,7	7,7
30	95 ± 5	6,2	9,5	7,4	7,0
	F	176,97 ***	63,42 ***	33,55 ***	19,98 ***
	LSD	0,6	1,4	1,0	1,3

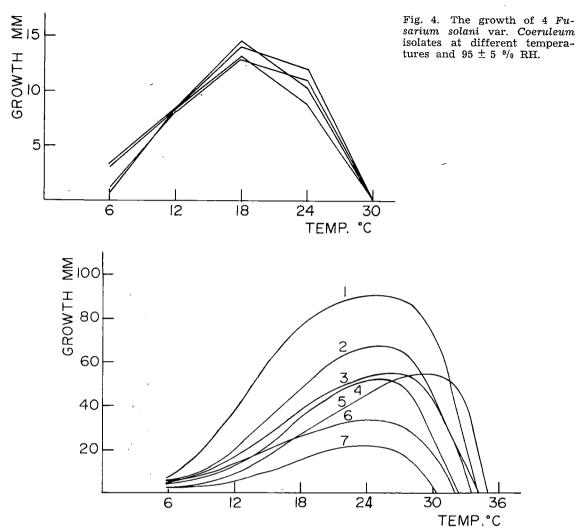


Fig. 5. The growth of some Fusarium spp. on PSA at different temperatures over 4 days. The fungi demonstrating almost the same growth rates are given in parenthesis. 1. Fusarium culmorum (F. graminearum). 2. F. sulphureum (F. trichothecioides, F. tricinctum, F. sambucinum, var. coeruleum). 3. F. sporotrichioides (F. oxysporum, F. sambucinum, F. solani). 4. F. avenaceum. 5. F. oxysporum var. redolens. 6. F. acuminatum. 7. F. solani var. coeruleum.

symptoms developed were almost the same at both optimum temperatures (Fig. 6).

Fusarium culmorum (W. G. Smith) Sacc. was like F. sporotrichioides in many respects. It is not a strong pathogen, and demonstrates two optimum temperatures at about 12°C and between 24 and 30°C, but it seemed to favour higher RH (Table 6 and Fig. 2). At higher temperatures it produced symptoms

of powdery rot.

Fusarium solani var. coeruleum Sacc. Booth comb. nov. is pathogenic at low temperatures like all the species named above but its maximum is lower (hardly 30°C), the optimum being nearby 20°C. However, it grows very well from 12 to 24°C (Table 7 and Fig. 2). The optimum temperature of this fungus and that of resistance reactions of the potato are

Table 7. The growth of Fusarium solani var. coeruleum (7572-5) under different environmental conditions. Incubated for 20 days.

	1		
1980 June	Axia:	1,0 1,0 	*** 230,39 *** 1,1
	Radial	1,0 1,0 1,0 6,9 7,9 13,5 13,5 10,9 10,9	88,05 *** 2;
November	Axial	2,55 - 2,51 8,44 - 1,59 - 2,94 - 2,94 - 3,96 - 3,96	77,02 *** 48,02 *** 0,5 1,1
Nove	Radial	1,2 1,2,1 1,4,1 1,4,1 1,4,	77,02 * 0,5
ây	Axial		** 54,74 *** 1,8
May	Radial	4,5   4,1	31,19 *** 1,7
April	Radial Axial	1,1 1,2 1,2 2,4 3,8 6,6 6,6 9,8 13,3 13,3	184,24 *** 56,34 *** 0,6 1,4
1979 January	dial Axial	1,0 2,3 1,2 2,0 2,4 1 2,1 1,1 1,7 1 1,2 1,1 1,1 1,1 1,2 1,2 1,2 1,2 1,2 1	28,82 *** 42,94 *** 0,8 1,3
p. RH.		50 50 50 50 50 50 50 50 50 50 50 50 50 5	F 28 LSD (
Temp.	ပ္	6 6 112 112 118 118 118 124 242	

presumably almost indentical, which has resulted in a wide optimum range (12—24°C) with no peak. The result was confirmed with several isolates (Fig. 4). This fungus clearly favoured the high RH, especially at higher temperatures (24°C). The symptoms which developed in each treatment were fairly similar, the cut surface usually light brown, rapidly becoming rust brown and later dark brown (Fig. 6).

Fusarium graminearum Schwabe was like F. s. v. coeruleum in many respects, but was stronger pathogen. It favoured a higher temperature (optimum over 20°C) (cf. Tivoli & Jouan 1980) and was completely dependent on high RH (Table 8 and Fig. 2). The symptoms resembled those produced by F. s. v. coeruleum but the rust brown phase was darker and the dead flesh rapidly became dark brown, nearly black (Fig. 6).

Fusarium avenaceum (Corda ex Fr.) Sacc. is a strong pathogen on potato tubers. It favoured conditions of high temperature and high RH. The optimum growth temperature was near 30°C (Table 9 and Fig. 2) and it was so highly dependent on the high RH (Table 10) that one or two weeks after inoculation, decreased RH greatly retarted its growth. The symptoms it produced under optimum conditions were rather profuse growth of white or red mycelia on the lesion, the cut surface being greyish brown, moist and rapidly darking; sometimes a cavity had developed at the centre of the lesion and was covered with reddish mycelia (Fig. 6).

Fusarium tricinctum (Corda) Sacc. is a weaker pathogen with optimum growth conditions of the high temperature (about  $30^{\circ}$ C) and high RH (95  $\pm$  5%). It also infects potato tubers at lower values of temperature and RH (Table 11 and Figs. 2 and 8).

Fusarium oxysporum Schlecht emend Snyder & Hansen, F. oxysporum var. redolens (Wollenw.) Gordon and F. solani (Mart.) Sacc.





19111

Fig. 7. The growth of F. sulphureum in tubers of cv. Bintje. The tubers were incubated for 20 days at temperatures of 6, 12, 18, 24 and 30°C and low RH. Note different symptoms developed at low and high temperatures.

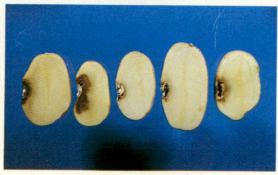


Fig. 6. The growth of Fusarium avenaceum, F. graminearum, F. solani var. coeruleum and F. sporotrichioides in tubers of cv. Bintje. The tubers were incubated for 20 days at temperatures of 6, 12, 18, 24 and 30°C (left to right) and 95  $\pm$  5 % RH.

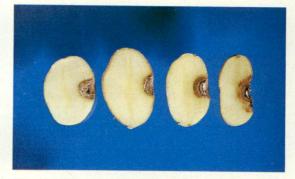


Fig. 8. The growth of Fusarium tricinctum in tubers of cv. Bintje, incubated for 20 days at temperatures of 12, 18, 24 and  $30^{\circ}$ C and high RH.

Table 8. The growth of Fusarium graminearum under different environmental conditions. Incubated for 20 days.

Temp		March	1980	Apr	il 1980
°C	%	Radial	Axial	Radial	Axial
6	95 ± 5	1,0	2,7	0,8	2,6
12	95 ± 5	9,1	10,7	9,7	9,4
18	95 ± 5	19,6	17,8	22,4	14,6
24	95 ± 5	14,5	16,5	27,8	16,3
30	95 ± 5	16,3	14,8	15,9	11,0
	F	44,71 ***	145,57 ***	281,25 ***	92,09 ***
	LSD	3,4	1,6	2,0	1,8

Table 9. The growth rate of Fusarium avenaceum (isolate 7760—1) at different temperatures. Incubated for 20 days at 95  $\pm$  5% RH.

		`— <u> </u>
er	Axial	4,4 7,5 9,6 10,6 12,8 2,2
October	Radial	2,5 4,3 6,1 8,6 11,4 12,8 18,43 *** 21,68 ***
۶	Axial	
May	Radial Axial	9,9   15,14 * 3,2
ril	Axial	21,9  20,2  4,09
April	Radial Axia	
<b>ਸੂ</b>	Axial	
March	Radial Axial	6,1   14,6   96,11 **
19 ary	Axial	2,5   3,9   5,8   6,1   12,0   12,0   12,0   6,1
1979 January	Radial Axia	2,5 5,8 6,1 12,0 — — — — — — 141,09 *
3 1ber	Axial	4,4 7,6 7,8 8,0 12,6 12,8 17,6 1.2,8 17,6 1.2,8 7.3,33 *** 15,12 * 2,8 4,0
1978 November	Radial	4,4 5,6 8,0 12,8 ————————————————————————————————————
Temp.	ာ့ '	6 12 18 18 30 8 1 1.SD5 %

emend Snyder & Hansen. We only carried out preliminary tests on these three fungi. According to the results (Table 12), they seemed to be quite weak pathogens with growth optima of high temperature and high RH like F. tricinctum. Further tests are needed to obtain more reliable results.

Fusarium acuminatum Ellis & Everthart and F. sambucinum Fuckel. The isolates of these two fungi in our tests were so weak that we obtained no values for their optimum growth conditions.

The approximate growth curves of the fungi on PSA are presented in Fig. 5. We can establish that only a few fungi with high temperature optima have almost the same growth curve on PSA and in potato tubers. This verifies the assumption presented above that living tubers as growth substrate probably influence the growth of the fungi in different ways at different temperatures.

The results presented above do not give complete information on the problem studied, but they give plenty of new facts about the optimum growth conditions of *Fusarium* species as pathogens of the potato. A larger number of different conditions must be used for more exact determination of optimum temperature and relative humidity. Only a few species reported as pathogens of potato tubers were not included in this study.

The aim of the study was to investigate the potential of thermotherapeutic control of Fusarium rots. This seems to be difficult to determine in the light of the results of this study because some common and strong pathogens may infect tubers under any conditions. One solution may be that of growing a cultivar which is fairly resistant to one or two pathogens or lack of certain pathogens in an area that makes possible the simpler control of other pathogens. Further trials are necessary to establish whether or not this would work.

Table 10. The effect of fluctuation in the RH of the incubation chamber on the growth of Fusarium avenaceum and F. sulphureum, the former incubated at 24°C and the latter at 12°C.

Incu	bating 1	RH			Fusarium av	enaceum		Fusarium sulphureu		
	ng weel	C.		May 1979		December	1979	May 1979		
	2. ———	3.		Radial	Axial	Radial	Axial	Radial	Axial	
95 95 95 50 50	95 95 50 50 50 95	95 50 50 50 95	•	19,0  6,9   5,3   1,4 1,6 1,8	21,6  11,3   7,7   3,1 3,7 4,3	14,0 13,6 10,6 1,0 1,2 1,5	16,7 16,5 14,7 1,7 2,6 3,0	24,8   24,1   24,2   26,2   24,6   23,2	17,7   15,0   18,4   20,1  15,4   14,7	
			F LSD	315,08 *** 1,6	339,34 *** 1,6	86,55 *** 2,1	93,06 ***	4,05 * 1,5	32,98 ***	

Table 11. The growth of  $Fusarium\ tricinctum\ under\ different\ environmental\ conditions.$  Incubated for 20 days.

Temp. RH	March	1980	April 1980		
°C %	Radial	Axial	Radial	Axial	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	1,5 0,9 1,0 1,5 	2,5     1,7 1,7 1,7   2,9   — 12,7	1,4 1,4 1,6 2,4 1,2 6,8	2,6 2,8 2,5 4,1 1,3 7,3	
F LSD	227,76 *** 1,2	316,76*** 0,9	49,13 ***	3,83 * 2,8	

Table 12. The growth of Fusarium oxysporum, F. oxysporum var. redolens and F. solani at different temperatures, RH at each temperature was 95  $\pm$ 5%. There was no growth in low RH (50  $\pm$  10) at a temperature of 24°C. Incubated for 20 days.

Temp.		the means of radial and axial growt	hs
°C	Fusarium oxysporum	F. o.v. redolens	F. solani
6 12 18 24 30	1,0 1,0 1,0 1,9 1,8	1,0 1,0 2,5 9,4	1,0 1,6 1,8 2,5
F LSD	11,46 ** 0,6	246,12 *** 0,7	8,01 ** 0,8

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#### SELOSTUS

#### Fusarium-sienten kasvun optimit Bintjen mukuloissa

#### ESKO SEPPÄNEN

#### Maatalouden tutkimuskeskus

Tutkimuksessa selvitettiin 14 perunalle patogeenisen Fusarium-lajin kasvunopeutta lämpötiloissa 6, 12, 18, 24, 30 ja 36°C sekä erilaisissa kosteusoloissa. Ravintoalustalla kasvatettu sieni inokuloitiin korkkiporalla mukulan keskivaiheille tehtyyn haavaan (2 mm syvä, 5 mm läpimitta), jonka jälkeen mukulat säilytettiin eri olosuhteissa 20 vrk. Sienten kasvu todettiin pitkittäin halkaistusta mukulasta mittaamalla keskimääräinen eteneminen sekä pinnansuuntaisesti että syvyyssuuntaan.

Tavoitteena oli selvittää phomamädän torjunnassa käytettävän lämpökäsittelyn soveltamismahdollisuudet *Fusarium*-sienten torjumiseksi. Eri *Fusarium*-lajien likimääräiset kasvun optimiolosuhteet olivat seuraavat:

Fusarium sambucinum Fuckel var. coeruleum Wollenw. suosi alhaista lämpötilaa ( $10-12^{\circ}$ C) ja alhaista ilman suhteellista kosteutta (RH  $40-50^{\circ}$ ).

F. sulphureum Schlecht. ja F. trichothecioides Wollenw. in Jamieson & Wollenw. niinikään suosivat alhaista kosteutta, mutta niillä oli kaksi optimilämpötilaa (10—12 ja 24—27°C).

F. culmorum (W. G. Smith) Sacc. ja F. sporotrichioides Sherb. Myös näillä sienillä oli kaksi optimilämpötilaa (10—12°C ja ylempi 27—30°C), mutta nämä molemmat, etenkin F. culmorum, suosivat suurta kosteutta (RH 90—100  $^{0}/_{0}$ ).

F. solani var. coeruleum Sacc. Booth comb. nov. ja F. graminearum Schwabe kasvoivat parhaiten n. 20°C:n lämpötilassa, jälkimmäisen optimi saattaa olla hiukan korkeampi ja vaativat, etenkin F. graminearum, korkeata ilman kosteutta.

F. avenaceum (Corda ex Fr.) Sacc. ja F. tricinctum (Corda) Sacc. suosivat korkeata lämpötilaa (n. 30°C) sekä suurta kosteutta.

F. oxysporum Schlecht. emend Snyder & Hansen, F. oxysporum var. redolens (Wollenw.) Gordon ja F. solani (Mart.) Sacc. emend Snyder & Hansen olivat mukana vain yhdessä kokeessa ja näyttävät suosivan korkeata lämpötilaa sekä suurta kosteutta.

F. acuminatum Ellis & Everhart ja F. sambucinum Fuckel. Näistä sienistä oli käytettävissä niin heikot isolaatit, että ne pystyivät vain tartuttamaan perunan, mutta ei etenemään siinä.

Vain muutaman sienen optimi kasvaessaan perunassa oli lähes sama kuin keinoalustalla. Syynä muutamien lajien kahteen optimilämpötilaan lienee perunan mukulan puolustusreaktioiden voimakkuus lämpötilassa 15—25°C.

Saatujen tulosten perusteella näyttävät mahdollisuudet soveltaa lämpökäsittelyä Fusarium-lajien torjuntaan vähäisiltä, koska melkein missä tahansa lämpötilassa 10—30°C jokin vahvoista perunan patogeeneista menestyy hyvin. Soveltamismahdollisuudet paranevat kuitenkin oleellisesti, jos tiedämme, että tietyllä alueella ei esiinny tiettyjä sienilajeja. Toisena mahdollisuutena on se, että viljelemämme perunalajike on kohtalaisen kestävä yhtä tai useampaa Fusarium-lajia vastaan. Näiden mahdollisuuksien tarkempi selvittäminen vaatii lisätutkimuksia.

#### Seria PHYTOPATHOLOGIA N. 82 — Sarja KASVITAUDIT n:0 82

# FUSARIUMS OF THE POTATO IN FINLAND III. VARIETAL RESISTANCE OF POTATO TUBERS TO SOME FUSARIUM SPECIES

#### ESKO SEPPÄNEN

Seppänen, E. 1981. Fusariums of the potato in Finland III. Varietal resistance of potato tubers to some Fusarium species. Ann. Agric. Fenn. 20: 177—183.

Tubers of 22 cultivars were tested for their physiological resistance to Fusarium sulphureum, F. avenaceum and F. solani var. coeruleum. Simple preliminary tests were also carried out to study their resistance to F. sporotrichioides, F. culmorum, F. graminearum and F. sambucinum var. coeruleum. All the tests were carried out under the optimum environmental conditions for each fungus.

The resistance of the cultivars varied widely, and was often quite different with different Fusarium species. Some cultivars seemed to have a fairly high resistance to all species tested. The old Eigenheimer and a new Finnish cultivar, Hankkijan Tuomas, were the most resistant, and also Sabina, Saturna and Jaakko possessed horizontal resistance to some degree. The results obtained with a particular fungus only occasionally correlated with those of other fungi.

Index words: Potato, varietal resistance, dry rot, Fusarium spp.

#### INTRODUCTION

The variation in the resistance of potato cultivars to Fusarium dry rot has long been known. In 1917, Pethybridge and Lafferty recognized that some cultivars were more resistant than others to Fusarium coeruleum (F. solani var coeruleum). Boyd (1952) studied the factors of resistance and divided them into the mechanical resistance of the tubers to wounding and the physiological resistance of the tissue to infection by fungi. He screened the resistance of a number of cultivars to both these and did not ascertain any inter

dependence between them. Moore (1945) recognized different degrees of resistance of cultivars to F. solani var. coeruleum and to F. avenaceum, and Ayers (1961) recognized different resistance to the former and to F. sulphureum. According to Langerfeld (1977) the incubating conditions and the length of the storage period before the test have different effects on the resistance of cultivars.

The aim of this study was to screen the physiological resistance of the cultivars grown in Finland to some *Fusarium* species.

#### MATERIAL AND METHODS

In 1979 17 and in 1980 22 cultivars were grown in sand soil, harvested and stored using identical, common practices. The latter season was especially favourable and even the latest cultivars such as Pito, Prevalent and Sanna started to mature. Tubers of nearly the same size were used for the tests. They were washed and dried well to prevent the natural infection caused by other fungi and bacteria.

The fungi used in the actual experiments were Fusarium sulphureum Schlecht., F. avenaceum (Corda ex Fr.) Sacc. and F. solani var. coeruleum Sacc. Booth comb. nov. and those used in the simple preliminary tests were F. culmorum (W. G. Smith) Sacc., F. graminearum Schwabe, F. sambucinum Fuckel var. coeruleum Wollenw. and F. sporotrichioides Sherb., a single isolate of each. Their isolation and identification, as well as their growth under different environmental conditions has been reported in earlier papers (Seppänen 1981 a, b). To maintain the pathogenicity of each fungus they were grown just before experiments in Bintje tubers, isolated conventionally and grown on potato sucrose agar (PSA). The age of the colonies used varied from 2 to 5 weeks and

was always during the period of profuse spore production.

The tubers were inoculated using an adaptation of the method of Langton (1971). A wound 5 mm in diameter and 2 mm deep was made with a cork borer, at the midpoint between the heel and rose end of the tubers. The mixture of pure cultures of the fungus, including the remaining agar, was used as inoculum. The wounds were filled with the inoculum and left uncovered. This method was equally as effective as plug infection or spore suspension infection. The experiments involved 20 tubers of each cultivar with 3 replicates. The preliminary tests presented in Table 4 involved only 10 tubers and no replicates. The tubers were incubated for 20 days under the optimum conditions for each fungus. For analysis, the tubers were halved longitudinally through the infection locus and the maximum radial and apical growths were measured. Their averages are presented here. All the results of the actual experiments were analyzed using variance analysis and the LSD values were calculated with Tukey-Hartley tables. Some correlations between separate tests were calculated, and the significances of these coefficients were tested with a t-test.

#### RESULTS AND DISCUSSION

Fusarium sulphureum being a strong pathogen, infected all the tubers easily and its advance in the tubers of most cultivars was fairly equal. The results of the tests carried out under similar conditions correlated highly significantly. The correlation between tests carried out at the lower and higher optimal temperatures of the fungus was weaker. Some cultivars which were fairly resistant

at the lower temperature were rather susceptible to infection at the higher one. This confirmed the result of Langerfeld (1977), that resistance reactions may vary according to temperature.

Cultivars Ostara and Jaakko were the most resistant at both temperatures (12 and 24°C), but, excluding the results obtained at 24°C, Sabina and Eigenheimer also belong

Table 1. The growth of Fusarium sulphureum in different cultivars during 20 days' incubation under different conditions of temp. and RH.

Cultivar	Nov. 1980	Dec. 1979	Dec. 1980
	12°C/60 ± 10 %	12°C/90 ± 5 %	24°C/50 ± 10 %
Sabina Ostara Eigenheimer Hankkijan Timo Sieglinde Jaakko Hankkijan Tuoma Sirtema Record Saturna Maris Piper Jo 0701 Prevalent Posmo Pito Provita Stina	12°C/60 ± 10 %  6,6 6,9 7,4 8,2 8,5 8,6	7,1   6,9	13,6     8,2   15,1       10,2   15,0       9,0   14,3     18,4       12,8   17,8   17,8   16,9       20,9   15,6     21,3   21,3   21,3
Olympia Barima Bintje Sanna Veto	12,2 12,4 13,5 13,7 13,9	11,3   11,6   11,6   12,5	13,1       12,7 13,8     19,3     14,7
F	126,08 ***	66,49 ***	16,50 ***
	SD <sub>5</sub> % 0,6	0,7	2,2

to the most resistant cultivars, followed by Hankkijan Timo, Sieglinde and Hankkijan Tuomas (Table 1).

The first reports available concerning the varietal resistance to *F. sulphureum* were presented by Wojciechowska-Kot (1975) and Czajka (1977). The former incubated the tubers at 15—17°C and 70—90 % RH. There were only 4 cultivars: Saturna, Bintje, Sieglinde and Sirtema, common to her and to our tests. The results we obtained at 24°C were similar to hers but the results we obtained at 12°C were not.

The recistance to *F. avenaceum* varied greatly (Table 2). The fungus advanced four time more in the tubers of the most susceptible cultivars than in the most resistant ones. Sabina, Provita and Veto were the most resistant in these tests, followed by Maris Piper, Hankkijan Tuomas, Stina and Jaakko. Bintje was one of the most susceptible cultivars, as it

Table 2. The growth of Fusarium avenaceum in 20 days at 24°C and RH 95  $\pm$  5 %. The percentage infection was 100 %.

Cultivar	Nov. 1980	Jan. 1980
Sabina	4,0	3,3
Provita	5,1	1,7
Veto	6,0	4,6
Maris Piper	7,4	<del></del> .
Hankkijan Tuomas	7,7	3,8
Stina	7,7	1,9
Jaakko	8,8	4,3
Posmo	9,4	_
Prevalent	9,6	<u> </u>
Sieglinde	10,0	8,2     7,8
Saturna	10,2	7,8
Eigenheimer	10,7	ll. —
Sirtema	11,4	[]] 8,3 [
Hankkijan Timo	12,1	[[[ 6,5 ]
Olympia	12,3	9,3
Record	13,1	7,7
Barima	13,1	
Bintje	13,4	13,0
Sanna	13,9	7,7
Pito	13,9	6,5
Ostara	14,5	—
Jo 0701	16,3	11,4
F	^66,35 ***	39,02 ***
${ m LSD}_5\%$	- Y	1,5

Table 3. The growth of F. solani v. coeruleum in 20 days at 18°C and RH 95  $\pm$  5%. The percentage infection was 100%.

Cultivar	Feb. 1981	Dec. 1979
Hankkijan Tuomas	3,1	1,6
Eigenheimer	3,7	-,-,
Sirtema	3,7	5,1
Sabina	4,9	3,4
Posmo	4,9	
Sanna	5,2	9,5
Saturna	6,3	6,6
Stina	6,4	6,0
Olympia	7,1	2,0
Prevalent	7,1	
Veto	7,5	3,2
Pito	8,2	3,3
Barima ,	8,4	<del></del>
Jo 0701	8,9	13,4
Jaakko	9,6	5,7
Bintje	9,6	9,8
Record	10,4	12,3
Ostara	10,5	12,0
Provita	11,4	<sup>'</sup>   8,9
Sieglinde	11,5	5,9
Hankkijan Timo	12,9	3,4
Maris Piper	13,8	'  '
F	105,24 ***	72,31 ***
$\mathrm{LSD}_{5\%}$	0,8	1,2

was to *F. sulphureum*. Ostara, which was fairly resistant to *F. sulphureum*, proved to be very susceptible to *F. avenaceum*. Despite some different results in these two tests with *F. avenaceum*, there was a high correlation between them.

There is no information in the literature concerning earlier tests of varietal resistance to *F. avenaceum*. Further tests under more practical conditions are needed.

The varietal resistance to F. solani var. coeruleum also varied greatly (Table 3), and the results obtained in these two tests deviated from each other more than in tests with F. sulphureum and F. avenaceum. Possible reasons for this might be that the tubers used in each test were grown in different seasons, the tests were carried out at different times during the storage period, and there might be a great deviation within some cultivars; e.g., in the tests with F. s. var. coe-

Table 4. Preliminary results on varietal resistance to 4 Fusarium species. The growth of the fungi in millimetres under incubating conditions ( ${}^{\circ}\text{C}/{}^{0}/_{0}$  RH) favourable to each fungus measured 20 days after cortical inoculation. The second column indicates the order of resistance (Most resistant = 1).

Cultivar	F. cult	morum	F. gram	inearum	F. samb		F. sporotr	ichioides
	12 ± 1,	/40 ± 5	$24\pm1/95\pm5$		$\begin{array}{c} \text{var. } coeruleum \\ 12 \pm 1/40 \pm 5 \end{array}$		$12\pm1/95\pm5$	
Barima	2,8	2	14,4	14	4,2	11	11.4	
Bintje	8,6	18	16,3	20	17,8	$\frac{11}{22}$	11,4	18
Eigenheimer	6,4	12	15,3	18	3,6	4	12,5	19
Hankkijan Timo	7,1	14	15,8	19	3,7	5	5,7	3
Hankkijan Tuomas	9,5	20	13,5	11	7,2	19	6,3	5 16
Jaakko	7,8	17	7,0	î	4,2	12	10,6	
Jo 0701	8,7	19	14,0	13	6,2	18	9,9	12
Maris Piper	3,7	7	10,7	4	5,9	16	8,6	10
Olympia	6,8	13	13,0	10	8,2	20	9,5	11
Ostara	3,4	4	12,2	7	4,0	20 9	15,1	22
Pito	6,2	11	12,4	8	3,9	9 7	4,2	1
Posmo	3,5	5	15,2	17	3,9	8	8,0	9
Prevalent	3,6	6	11,4	5		-	7,9	8
Provita	3,1	3	7,1	2	4,8	15	10,0	13
Record	2,6	1	15,0	16	4,1	. 10	10,4	14
Sabina	$\frac{2,0}{7,2}$	15		21	3,8	6	4,9	2
Sanna	5,9	10	16,4	22	5,9	17	6,3	6
Saturna	4,9	9	16,4	22 15	3,5	3	11,0	17
Sieglinde	7,4	16	14,6		3,0	1	6,2	4
Sirtema	9,6	21	12,7	9	3,3	2	7,8	7
Stina	3,9	8	13,8	12	4,3	13	12,5	. 20
Veto	11,4		8,4	3	4,4	14	10,5	15
		22	11,5	6	12,5	21	14,1	21

ruleum the infection was progressive in some of the tubers and the growth of the fungus after infection was arrested by the resistance of the other tubers.

Hankkijan Tuomas, Eigenheimer and Sirtema were the most resistant, followed by Sabina and Posmo. Maris Piper seemed to be very susceptible, and Bintje, Record, Ostara, Provita, Sieglinde, Jaakko and Hankkijan Timo were considered susceptible. The resistance of some cultivars was aproximately the same as ascertained by Boyd (1952), Kranz (1959), Wojciechowska-Kot 1975, Bång (1976) and Langerfeld (1977), but only some of the cultivars were common to this and any of the other investigations. Further tests are needed for a more reliable picture of the resistance of certain cultivars.

The results of preliminary tests with F. culmorum, F. graminearum, F. sambucinum var. coeruleum and F. sporotrichioides are presented in Table 4. These fungi, although strong pathogens in some cultivars, . are not as common as the three presented earlier. On the other hand, these first tests were carried out without replicates and can hardly give a reliable picture of the resistance of the tested cultivars. However, we can again ascertain a great variation within and between the fungi. Ostara, Record, Provita and Saturna seemed to have a certain amount of horizontal resistance to these pathogens, and Bintje, Hankkijan Tuomas, Olympia, Sirtema and Veto demonstrated clear susceptibility.

In these tests, no cultivar proved to be equally resistant to all the fungi tested. Cultivars Eigenheimer, Hankkijan Tuomas, Sabina, Saturna and Jaakko showed fairly good, horizontal resistance to the three dominant pathogens (Tables 1—3), but seemed to be very susceptible to some of the others. Bintje was the only cultivar which was very susceptible to infection by all the fungi used as pathogen, whereas many other cultivars with

Table 5. The correlation coefficients between the results of separate tests.

	1	2	က	4	ທ	9	7	8	6	10
1 Fusarium avenaceum Jan80										
avenaceum Nov80	0,82***									
F. sulphureum Dec79 (12°)	0,25	0,24								
1 (12°)	0,22	0,12	0,91**							
$(24^{\circ})$	0,27	0,19	0.53*					•		
solani v. coeruleum Dec79	0,42	0,46	90,0		0,24		`			
solani v. coeruleum Feb81	0,14	0,12	-0,24	1	-0,36	0,33				
sporotrichioides Dec80	- 60'0	-0,13	0,71**		0,26	-0,26	-0.18			
-81	*09.0	0,42	-0,03	1	0,20	0,10	0,32	-0.24		
F. culmorum March -81	0,25	-0,05	0,16	0,04	0,12	-0,37	-0,28	0,41	0,16	
F. sambucinum v. coeruleum M81	0,37	-0,04	0,45		-0,03	-0,01	0,03	0,55**	0,57**	0,52*

clear susceptibility were fairly resistant to one or more fungi.

The results presented here indicate only the physiological resistance of the cultivars. In practice, the other component (mechanical resistance) is as important, and so a cultivar such as Record, which shows good resistance to wounding although it is susceptible to many fungi, can be grown successfully.

The correlation coefficients between the results of the tests are quite variable (Table 5). Excluding correlations between tests with the same fungus, there seem to be positive correlations between

F. avenaceum and F. graminearum

F. sulphureum » F. sporotrichioides

F. sulphureum » F. sambucinum var. coeruleum

F. sporotrichioides » F. sambucinum var. coeruleum

F. culmorum » F. sambucinum var. coeruleum

The results clearly indicate the disimilarity between the different fungi. However, the size of the material examined does not permit final conclusions to be drawn.

The results presented concern the varietal resistance under conditions most favourable to each fungus. The results obtained at 12 and 18°C will be adapted directly to corre-

spond to the wound healing period just after harvest, during which most infections occur. Further tests will be carried out at lower temperatures to extend our knowledge of the variation in varietal resistance and to find possible methods for the control of Fusarium dry rots during the wound healing period.

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#### SELOSTUS

#### Perunalajikkeiden kestävyys Fusarium-sieniä vastaan

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#### Maatalouden tutkimuskeskus

Maassamme viljeltävien perunalajikkeiden kestävyyttä eri Fusarium-sieniä vastaan tutkittiin tartuttamalla mukulat niihin tehtyihin haavoihin ravintoalustalla kasvatetun puhdasviljelyksen seoksella (rihmasto + itiöt + jäljellä oleva agar). Tartutuksen jälkeen perunat säilytettiin 3 viikkoa kullekin sienelle suotuisissa oloissa. Perusteellisemman tutkimuksen kohteena olivat merkittävimmät patogeenit Fusarium sulphureum, F. avenaceum ja F. solani var. coeruleum, alustavasti selvitettiin lajikekestävyyttä myös F. culmorumia, F. graminearumia, F. sambucinum var. coeruleumia ja F. sporotrichioidesta vastaan.

Lajikkeiden kestävyys eri Fusarium-lajeja vastaan oli hyvin vaihteleva. Monet näyttävät omaavan kohtalaisen kestävyyden joitakin lajeja kohtaan, mutta ovat hyvinkin alttiita muita vastaan.

Vain muutamat lajikkeet näyttävät omaavan tyydyttävän yleiskestävyyden kolmea merkittävintä patogeenia vastaan, mutta eivät kaikkia muita vastaan. Mainittavaa yleiskestävyyttä todettiin lajikkeilla Eigenheimer ja Hankkijan Tuomas, seuraavina olivat Sabina, Saturna ja Jaakko. Bintje oli altis kaikille tutkituille sienille. Myös Rekord osoittautui yllättävän heikoksi. Nämä tulokset osoittavat vain lajikkeiden sienenkestävyyttä ja taudinkestävyyteen kuuluu lisäksi käsittelynkestävyys. Rekord omaa muita paremman käsittelynkestävyyden ja senvuoksi sitä voidaan pitää yleiskestävyydeltään tyydyttävänä.

Verrattaessa lajikkeiden alttiutta eri sienilajeja kohtaan todettiin, että sienilajien patogeenisuus on hyvin spesifinen. Vain muutamien lajien kesken todettiin merkitsevä korrelaatio.

#### Seria PHYTOPATHOLOGIA N. 83 — Sarja KASVITAUDIT n:o 83

#### A REVIEW

#### START OF SEED POTATO PRODUCTION IN FINLAND

#### ERKKO PIETARINEN and ESKO SEPPÄNEN

Pietarinen, E. & Seppänen, E. 1981. Start of seed potato production in Finland. Ann. Agric. Fenn. 20: 184—187. (Agric Res. Centre, Seed Potato Centre, SF-91900, Liminka, Finland).

Modern Finnish seed potato production originates from the 1970s. The first nucleo stocks were produced from cvs. Pito and Tammiston Aikainen in 1971. Since then meristem cultured plants have been produced from 17 cultivars grown in Finland and from a number of breeders' selections.

Effective seed potato production started in 1976 when the Seed Potato Centre was founded. Production has increased evenly and in the 1981 nearly 4000 tons of seed potatoes comprising 12 cultivars were put on the market. Of this, 900 tons was from 6 cultivars based on meristem cultured stocks, 750 tons from 2 cultivars derived from clonal selected stocks, and 2250 tons from 4 cultivars originated from earlier imports.

Index words: Potato, meristem culture, cutting propagation, seed potato production.

The meristem culture method to produce virus-free potato plants from cultivars totally infected by one or more viruses has been used since the 1950s (Morel and Martin 1955). At the Institute of Plant Pathology, in Tikkurila, Tapio (1972) produced the first virus-free plants from two Finnish cvs. Pito and Tammiston Aikainen in 1971 and during the following two years from cvs. Amyla, Jaakko, Teho and Veto (Table 1).

The results of the propagation of first nucleo stocks were modest from the point of view of practical cropping. This situation changed with the founding of the Seed Potato Centre in 1976.

This paper deals with the cultivars from which nucleo plants have been produced, and the development of seed potato production between 1976 and '81.

#### **METHODS**

The meristem cultures were produced at the Institute of Plant Pathology using conven-

tional methods. The young plants were grown at temperatures of  $36-37^{\circ}C$  for 3-4 weeks

Table 1. The number of nucleo stocks (cultivars and breeders' selections) from which virus-free plants were produced during the period 1971—81. Cultivar names are given only in connection with the first year of production.

Year	Number of stocks	x, x 3	Names of cvs.	
1971	2		Pito, Tammiston Aikainen	
1972	4		Jaakko, Teho, Veto	
1973	4		Amyla	
1974	3		Breeder's selections only	
1975	3		Hankkijan Timo, Hankkijan Tuomas	
1976	6		Hankkijan Tanu (as a breeders' selection)	
1977	4		Breeder's selections only	
1978	5		Sanna, Sieglinde	
1979	5		Mandel, Olympia, Rosafolia	
1980	4		Record	
1981	8		Barima, Posmo	

before the removal of meristem tissues. The agar used was a slight modification of Murashige & Skoog's medium (Tapio 1972, Bremer and Korhonen 1978). After removal, the meristem tissue was grown at 24°C and in continuous light of 3000-4000 lux. Preliminary virus tests were made as the cultures reached a suitable size. PVX, PVS and PVM were tested for using a micro-agglutination method, PVA and PVY with A 6 leaflets. The tests were repeated about once a month. Some sap preparations from nucleo plants were studied with an electron microscope. Most problems in the production of virus-free plants were connected with PVS. However, in our experience the success of this method depends more on the cultivar than on the virus. Because of recontamination by bacteria and fungi, most cultivars must be meristem cultured regularily.

The propagation of healthy nucleo plants was carried out at the Seed Potato Centre, founded in the Tyrnävä and Liminka region in northern Finland in 1976. From 1976 to 1979, the first stage propagation of nucleo stocks based partly on the use of stem cuttings, and partly on the conventional use of tubers. Since 1980, the system has been

changed so that the first stage propagation is always based on stem cuttings. This turned out to be a simple and rapid method for propagating new meristem stocks. It also reduces the need for virus testing, and gives better control of bacterial and fungal diseases. During propagation, new stocks were regularly tested for viruses. Agglutination tests were used for PVX, PVS, and PVM. PVY was tested for using *Nicotiana glutinosa*, a test plant which also shows clear symptoms with PVX. The Elisa-test was taken into use for PVX, PVS, PVM, and PVY in 1981. Latent phoma infection was tested with the



Fig. 1. Meristem culture nucleo stocks were propagated in green houses from stem cuttings.

knack-test since 1980 and ring rot using the eggplant Solanum melongena since 1981.

During the first years, most attention was paid to elite seed production for commercial purposes. Production happened on private farms around the Seed Potato Centre, mainly in the parishes of Tyrnävä and Liminka.

Most stocks originated from imported seed, but in the production of certain cultivars clonal selection method was also used. The proportion of production originating from meristem cultures produced in Finland has increased each year.

#### SEED PRODUCTION

During a period of 10 years, healthy nucleo plants were produced from 17 cultivars and a number of breeders' selections (Table 1). Some of them have already reached large-scale production, some have had no practical use. Very much attention has been paid to new Finnish cultivars. The cropping of Finnish cultivars Pito and Jaakko, which had suffered a great set-back because of lack of good seed, has increased again.

Elite seed production for commercial pur-

poses has increased uniformly from 35 hectares in 1976 to 210 hectares in 1981. The quantity of seed production based on meristem culture in 1980 was nearly 900 tons, about 23% of the total production (Table 2). The quantity of seed derived from clonal selected stocks at the same time was 750 tons (19%) and the quantity of seed based on earlier imported seed 2250 tons (58%). Meristem cultured seed use is, however, increasing rap-

Table 2. Cultivars, number of nucleo- and breeders' seed stocks and the total seed production of each cultivar in 1980.

	Number of nucleo stocks produced by meristem culture and cutting propagation	Number of breeders' seed stocks	Total seed production (t)
Early cvs.			
Ostara	3	4	165
Hankkijan Timo	3	4	165 65 <sup>1</sup>
Sieglinde	4	2	$12^{1}$
Table potato cvs.	-	4	12-
Record	10	10	4 500
Pito	5	10 5	1 566
Sabina	<u> </u>	<b>.</b>	462 <sup>1</sup>
Jaakko	3	<del>-</del>	371
Hankkijan Tuomas	4	3	2261
Sanna	2	ა 1	50 <sup>1</sup>
Hankkijan Tanu	2	1	35 <sup>1</sup>
Olympia	7		$35^{1}$
	· · · · · · · · · · · · · · · · · · ·		· —
Cvs. for industrial use	_		
Saturna	5	3	$728^{2}$
Bintje	<del></del>	_	162
Frila	<del>-</del>		$35^{2}$
Eigenheimer	4	<del></del>	_
Posmo	3	_	_
Total	53	32	3 912

<sup>1 =</sup> meristem cultured origin

<sup>&</sup>lt;sup>2</sup> = clonal selected origin

idly and by the middle of the 1980s nearly all production will be cultivated from meristem cultured and cutting propagated origin.

The greatest problems in elite seed production was mechanical wounds because, as a result of the short growth period, tubers are

rarely matured when lifted. However, by using pre-sprouting, haulm defoliation and careful lifting the damage can be kept at an insignificant level. Virus diseases, late blight and even storage diseases were mostly easily controlled.

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#### SELOSTUS

#### Siemenperunan tuotanto

#### ERKKO PIETARINEN and ESKO SEPPÄNEN

#### Maatalouden tutkimuskeskus

Siemenperunan tuotanto sen nykyaikaisessa mielessä käynnistyi maassamme 1970-luvulla. Viruksettomien ydinkasvien tuotanto alkoi jo 1970-luvun alussa, jolloin kasvitautien tutkimuslaitoksella Tikkurilassa saatiin terveet ydinkasvit virustautien kokonaan tartuttamista lajikkeistamme Pidosta ja Tammiston aikaisesta (Tapio 1972). Työ on jatkunut kymmenisen vuotta ja tähän mennessä kaikkiaan 17 lajikkeesta ja useista jalostuslinjoista on tuotettu terve kanta-aineisto.

Terveen kanta-aineiston lisäys käytännön tarpeita varten käynnistyi 1976, jolloin perustettiin Siemenperunakeskus Tyrnävän-Limingan alueelle. Kanta-aineistojen pistokaslisäys kasvihuoneissa samoin kuin varsinainen siementuotanto on tapahtunut kokonaan siellä.

Toiminnan ensimmäisinä vuosina painopiste oli ulkomaista alkuperää olleiden kantasiemenerien lisäämisessä valiosiemeneksi. Tuotantoala on kasvanut vuosina 1976-81 35 hehtaarista 210 hehtaariin. Vuonna 1980 tuotetusta siemenestä noin 900 tonnia (23 %) polveutui kotimaisista kasvusolukkomenetelmällä tuotetuista kanta-aineistoista, 750 tonnia (19 %) kloonivalinnalla perustettuun kantasiemeneen ja 2250 tonnia (58 %) ulkomaiseen kantasiemenaineistoon. Kun toiminnan ensimmäisenä vuotena tuotanto perustui miltei yksinomaan ulkomaiseen kantasiemeneen, on nyt kasvusolukkoaineiston osuus lähes neljännes ja 1980-luvun puolivälissä päästänee miltei yksinomaiseen kotimaassa kasvusolukkomenetelmällä tuotetun kantasiemenen käyttöön.

#### Seria PHYTOPATHOLOGIA N. 84 — Sarja KASVITAUDIT n:o 84

#### RESEACH NOTE

# WITCHES' BROOM DISEASE OF ARCTOSTAPHYLOS AND VACCINIUM SPECIES IN FINLAND

#### KATRI BREMER

Bremer, K. 1981. Witches' broom disease of Arctostaphylos and Vaccinium species in Finland. Ann. Agric. Fenn. (Agric. Res. Centre, Inst. Pl. Path., SF-01300 Vantaa 30, Finland.)

Heavely stunted and abundantly branched Arctostaphylos uva-ursi, Vaccinium myrtillus and V. vitis-idaea plants with tiny leaves were found in forests in a few places in South and Middle-Finland in 1978. The symptoms were similar in all diseased plants.

Electron microscope observations on ultra-thin sections of eponembedded leaf tissues of *V. myrtillus* and *V. vitis-idaea* showed bodies similar to those of mycoplasma-like organisms. According to the diseased plants in the herbarium of the Helsinki University, the disease has occurred since 1910 in some places all over the Finland. This is the first record of the witches' broom disease in Finland.

Index words: witches' broom disease, mycoplasma, Arctostaphylos uvaursi, Vaccinium myrtillus, V. vitis-idaea.

A few strongly dwarfed and excessively branched blueberry *Vaccinium myrtillus* L. plants were brought to the author by Mr O. Heikinheimo (Agric. Res. Centre, Inst. Pest. Inv.) from Janakkala near Hämeenlinna in South-Finland. Later the same disease was observed by the author in plants of *Arctostaphylos uva-ursi* (L) Spr., *V. myrtillus* and *V. vitis-idaea* L. in forest in Porkkala near Helsinki.

Later diseased blueberry plants were found also in the vicinity of Jyväskylä in the middle of Finland (Raatikainen, M., oral communication).

The diseased plants of Arctostaphylos and Vaccinium species had tiny, light green leaves. The abundant erect branching gave

the plants a broom-like form. Some branches were dead (Fig. 1). The diseased plants did not produce any berries. Diseased plants in Porkkala occured in small batches on the rocky hills near the sea-side. The vegetation of the hills consisted mainly of shrubs Arctostaphylos, Calluna and Vaccinium and sparse wood of Pinus sylvestris and Picea excelsa.

Some experiments were carried out to confirm the cause of the disease.

Attempts to graft-transmit the disease agent from blueberry and bilberry (V. myrtillus and V. vitis-idaea) plants to the blueberry and bilberry seedlings and to high-bush blueberries (V. corymbosum L) failed. The grafted twigs did not grow together.

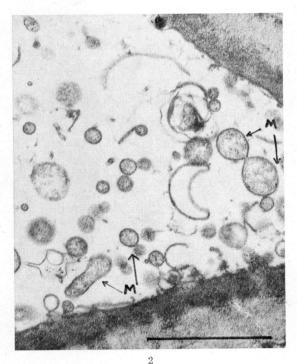


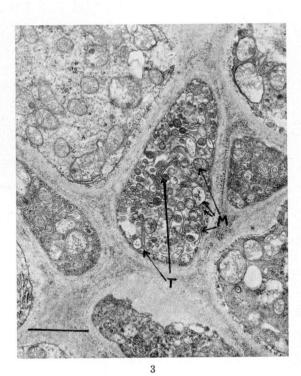
Fig. 1. V. myrtillus plants on the right are naturally infected with the witches' broom disease, plants on the left healthy.

In addition an electron microscopical study was carried out. Diseased and healthy looking blueberry and bilberry plants were collected in September from a forest in Porkkala.

Small pieces of the main veins of leaves were prefixed with  $2.5\,^{6}/_{0}$  glutaraldehyde in  $0.2\,$  M sodium phosphate buffer (ph 7.2) for  $2\,$  h in  $+5\,^{\circ}$ C. Then the tissue pieces were washed in the same buffer with  $0.2\,$  M sucrose, postfixed for  $2\,$  h with  $1\,^{6}/_{0}$  osmium tetroxide in  $0.1\,$  M sodium phosphate buffer (pH 7.2). The sections were obtained from Epon  $812\,$  embedded samples and stained with uranyl acetate and lead citrate. The sections were examined with the transmission electron microscope, Jeol  $100S\,$  operating at  $80\,$  kV.

Numerous bodies of the mycoplasma-like organisms were observed in cells of diseased blueberry and bilberry plants but not in the cells of healthy looking plants. The bodies were round or oval and their diameter varied from 50 to 1100 nm, many had a diameter of 100—200 nm (Figs. 2—4). Long, flexuous threads were found to occur in the same cells of blueberry as the mycoplasma-like organisms (Fig. 2), but not in cells of healthy plants. Those threads seemed to be of similar





Figs. 2—3. Electron micrographs of mycoplasmalike bodies (M) and long flexuous threads (T) in the cells of V. myrtillus. Bar represents 1  $\mu$ m.



Fig. 4. Mycoplasma-like bodies in the cells of  $V.\ vitis\mbox{-}idaea.$  Bar represents 1  $\mu m$ .

material as the membrane of the mycoplasmalike bodies. They might be membranes of degenerated bodies.

The witches' broom disease of Arctostaphylos and Vaccinium species seems to be similar to the witches' broom disease which has been found in V. myrtillus, V. oxycoccus, V. uliginosus, V. vitis-idaea and in Calluna vulgaris in Czechoslovakia (Blattný 1956, Blattny 1970), in V. myrtillus in the Netherlands (Bos 1960), in Germany (Uschdraweit 1961, Kegler et al. 1973) and in Yugoslavia (Blattný 1964).

In Czechoslovakia the disease was trans-

mitted by the leafhopper, *Idiodonus cruentatus* Panz (Blattný 1964).

Similar cells of mycoplasma-like organisms as in the present work have been found in diseased blueberry plants by Kegler et al. (1973), in the underground stems and roots of blueberry plants by Blattný and Vana (1974) and in blueberry stems by de Leeuw (1975).

The witches' broom disease seems to have occurred for a long time in Finland at least in blueberries but it was not recognized as a disease.

Hiitonen (1933) mentions a forma of V. myrtillus by name V. myrtillus microphyllum in the south-eastern parts of Finland.

In the herbarium of the Institute of Botany, University of Helsinki blueberry plants named *V. myrtillus microphylla* were seen by the author and recognized to be diseased by the witches' broom disease. These plants were collected from different places in Finland including Lapland. The oldest plants were collected in the year 1910. Thus this disease seems to be indigenous in Finland.

Acknowledgements. — The electron microscopy was carried out at the Department of Electron Microscopy, University of Helsinki, and I wish to express my sincere thanks to the personnel of this institute.

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#### SELOSTUS

### Mykoplasman aiheuttamaa pieniversoisuustautia mustikassa, puolukassa ja sianpuolukassa

#### KATRI BREMER

#### Maatalouden tutkimuskeskus

Kääpiökasvuisia, pienioksaisiksi tiheään haaroittuneita, hyvin pienilehtisiä mustikka-, puolukkaja sianpuolukkakasveja (kuva 1) löydettiin v. 1978 eräistä Etelä-Suomen metsistä. Kasveja epäiltiin sairaiksi.

Ymppäämällä ei onnistuttu siirtämään sairautta mustikoista ja puolukoista terveisiin mustikan, puolukan ja amerikkalaisen pensasmustikan siementaimiin.

Metsästä otetuissa sairaissa mustikoissa ja puolukoissa todettiin elektronimikroskoopin avulla mykoplasmojen kaltaisia organismeja (kuvat 3—5), joita ei löydetty terveen näköisistä kasveista.

Eräissä Keski-Euroopan maissa on esiintynyt mustikassa ja sen sukulaiskasveissa samanlaista pieniversoisuustautia (witches' broom disease, Hexenbesen-krankheit). Sairaista kasveista on löydetty mykoplasmojen kaltaisia organismeja. Meillä todettu tauti lienee samaa tautia.

Pieniversoisuustautia on todennäköisesti esiintynyt meillä jo kauan, joskin sitä ei ole aikaisemmin tunnistettu. Hiitonen on esittänyt v. 1933 julkaistussa kasviossaan mustikasta muodon Vaccinium myrtillus, forma microphyllum, joka kuitenkin on ominaisuuksiltaan mykoplasman tartuttaman mustikan kaltainen. Helsingin yliopiston kasvimuseon kokoelmissa on eri puolilta Suomea kerättyjen mustikkanäytteiden joukossa muutamia tautisia mustikkayksilöitä. Löytöjä on myös Lapista. Vanhin talletettu tautinen kasvi on vuodelta 1910.

#### Seria PHYTOPATHOLOGIA N. 85 — Saria KASVITAUDIT n:o 85

#### STRAWBERRY ROOT ROT IN FINLAND

#### PÄIVI PARIKKA

Parikka, P. 1981. Strawberry root rot in Finland. Ann. Agric. Fenn. 20: 192—197. (Agric. Res. Centre, Inst. Pl. Path. SF-01300 Vantaa 30, Finland).

A total of 486 strawberry plant samples from 153 plantations were investigated in 1978—1980. They originated from 29 localities in southern and central Finland. Plant roots were examined by moist chamber and agar plating methods. The fungal flora of the roots comprised 73 species. The most common pathogens were Fusarium fungi (8 species) including Fusarium avenaccum (Corda ex Fr.) Sacc., F. oxysporum Schlecht. and F. sambucinum Fuckel. Cylindrocarpon destructans (Zinssm.) Scholten and Botrytis cinerea Pers. ex Fr. were also prevalent. In addition, Coniothyrium fuckelii Sacc., Hainesia lythri (Desm.) Höhn. and Armillaria mellea (Vahl) Karst. were rather common. Gnomonia fragariae (Klebahn) and Coniothyrium fragariae Oudem. were rare but worth noticing.

Index words: Strawberry, root rot, Fusarium, Cylindrocarpon destructans, Gnomonia fragariae, Corniothyrium fragariae, Hainesia lythri, Armillaria mellea.

#### INTRODUCTION

The strawberry cultivation area in Finland has increased during the last ten years from nearly 1300 hectares in 1969 to about 2500 hectares in 1979. The increase has been particularly pronounced in central and eastern Finland. The most important strawberry cultivation areas are in North Savo and Varsinais-Suomi (Anon. 1969, 1979).

With the increase in cultivation area, specialised production and monoculture have also become common cultural practices. The adverse effects of monoculture on strawber-

ry plantations have led to increasing disease and pest problems. The importance of root rot in strawberry cultivation was first noticed in the 1960s and occurrence of common root rotting fungi was reported by Ylimäki (1970).

A combined study on strawberry root rots and root nematodes was begun in the Agricultural Research Centre in 1978. The aim of this study is to determine the fungal flora of strawberry roots and to discover the most common pathogens in the strawberry root rot complex.

#### MATERIAL AND METHODS

The material for this study comprised 486 strawberry samples collected from 153 plantations in 29 localities (Fig. 1). Of the samples, 230 originated from North Savo and 135 from Varsinais-Suomi. The total strawberry cultivation area on the plantations examined was about 300 hectares. They were mainly situated on moraine and fine sand, and also on clay soils in Varsinais-Suomi. The most common varieties cultivated were Senga Sengana and Zefyr. The ages

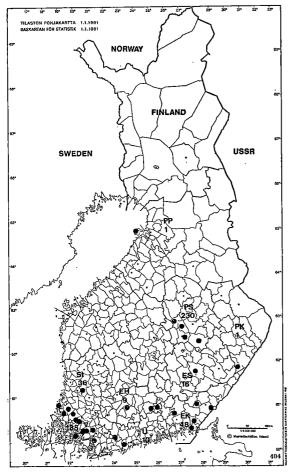


Fig. 1. The origin of strawberry samples by localities. Number of samples by biological provinces.

of plantations varied between 0,5 and 18 years and the average was two to six years (Fig. 2). Plants of low vitality or with various forms of damage were taken as samples.

The material was collected mainly during the summer months: in July in 1978, August-September in 1979 and July-August in 1980.

The plants were rinsed with running water and examined visually. The condition of the plant, colour of leaves, condition of roots and degree of rotting in the taproot were determined.

Leaves and roots were then removed and the taproot tissue was cut into pieces. Rootinhabiting fungi were cultivated by incubating the pieces on moist filter papers in Petri dishes. They were first kept for two weeks at room temperature ( $+20-+24^{\circ}$ C), then four weeks at  $+10^{\circ}$ C and again two weeks at room temperature. The culture period was thus at least two months. Fungi were also cultured on agar plates after cutting the taproot tissue into small pieces, removing the

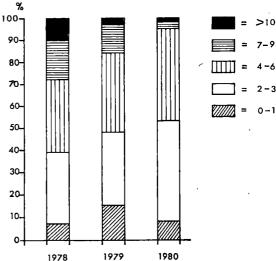


Fig. 2. The age of strawberry plants in percentages of the samples examined.

cortical layer and surface-sterilizing. The pieces were placed on Difco PDA (potato dextrose agar) and MA (malt extract agar) plates and incubated for two weeks at room temperature and then two weeks at  $+10^{\circ}$ C.

During the incubation period the dishes

and plates were occasionally examined with a stereomicroscope. The fungi were also identified by light microscope and then microphotographed by Dr. Kaiho Mäkelä. Pathogenic fungi were isolated and cultured on agar media.

#### RESULTS AND DISCUSSION

The plants examined were mainly in a moderately poor condition. Thirty to forty per cent of the plants were stunted and only 10 % in good condition. Some degree of root rot was found in nearly every plant. Severe root rot was found in a third of the taproots examined (Table 1). Only 5 % of the strawberry plants were healthy. The specimens were collected in order to find root -rotting fungi, and thus consisted of older plants, in which rots were most likely to appear (Mason and Rath 1978).

The most common fungi inhabiting strawberry roots were Mucor spp., Penicillium spp., Trichoderma spp., Gliocladium spp., Rhizopus, Fusarium spp., Cylindrocarpon spp. and Botrytis cinerea. A total of 73 fungus species was determined.

The most common pathogens found are named in Table 2. Fusarium spp., Cylindrocarpon spp., Rhizoctonia solani and Botrytis cinerea were common pathogens in rotten strawberry roots (Kudela and Sychrová 1978). In a former study, Ylimäki (1970) also found

these fungi prevalent in strawberry taproots, Fusarium species being the most common fungi isolated.

Fusarium species were present in nearly two-thirds (62  $^{0}$ /<sub>0</sub>) of the samples investigated in 1978—1980. They were also found in healthy-looking roots (Gourley 1969).

Fusarium avenaceum was the most common Fusarium species isolated. It can cause internal necrosis in strawberry roots (Jarvis and Hargreaves 1973).

F. oxysporum was not common in the material. The fungus is, however, reported to be a common root rot agent, as is F. culmorum (Łacicowa 1977). F. culmorum was found only twice in this study.

Cylindrocarpon species were found in one-third (32%) of the samples. Of these, C. destructans is pathogenic on strawberry taproots (Ylimäki 1970) and in the crowns of young plants (Montgomerie 1970). C. destructans was often found in rotted taproot tissues in this material.

Botrytis cinerea was also found to be pre-

Table 1. Different injuries in strawberry plants % of samples examined.

	Cor	nditior	ofp	lant	Co	plour of leaves Condition of roots			f roots	Degree of rot in taproot				
Year	Good	Moderate	Stunted	Dead	Dark green	Green	Reddish	Yellowish	Good	Moderate	Poor	Healthy	Slightly rotted	Rotted
1979 1980	13 10	54 47	31 41	3	5 3	78 58	22 37	4 3	11 14	65 64	23 22	11	66 63	24 40

Table 2. Occurrence of pathogenic fungi in strawberry roots percentages of samples examined.

	in roo	ungi % ts exar Year	examined		
Fungi	1978		1980		
·	No. of sa 128	mples 178	examined 180		
Armillaria mellea (Vahl)					
Karst.	4,7	7,3	7,8		
Botrytis cinerea Pers. ex Fr.	28,1	20,2	30,0		
Coniothyrium fuckelii Sacc.	3,1	10,1	10,6		
C. fragariae Oudem.	1,6	4,5			
Cylindrocarpon spp.	10,9	12,9	11,1		
C. destructans (Zinssm.)	,-	,-	•		
Scholten	26,6	23,6	13,3		
Diplocarpon earliana	•				
(Ell. & Ev.)	5,5	0	$^{2,2}$		
Fusarium spp.	23,4	27,4	14,4		
F. avenaceum (Corda					
ex Fr.) Sacc.	30,5	24,0	27,2		
F. culmorum (W. G. Smith)					
Sacc.	0	0,6	$^{0,6}$		
F. equiseti (Corda) Sacc.	<sub>;</sub> 0,8	0	0,6		
F. graminearum Schwabe	. 0	0,6	0		
F. oxysporum Schlecht.	2,3	$^{3,9}$	$^{3,3}$		
F. oxysporum var redolens	_				
(Wr.) Gordon	0	0,6	0,6		
F.poae (Peck) Wollenw.			0.0		
in Lewis	0	0	0,6		
F. sambucinum Fuckel	4,7	2,8	6,7		
Gnomonia fragariae	0.0	0.0			
Klebahn	3,9	0,6	$^{2,2}$		
Hainesia lythri (Desm.)	10.4	100	111		
Höhn.	16,4	16,3	11,1		
Phoma spp.	3,9	1,7	6,7		
Rhizoctonia solani Kühn	9,4	1,1 0	8,3		
Verticillium spp.	2,3		1,1		

valent (26 %) in the underground parts of strawberries. Hainesia lythri was present in rotted taproots alone or with other pathogens. The fungus is mentioned as root-rot pathogen by Strong and Strong (1931). H. lythri was also found in dead flower stalks. It formed profuse, light brown sporodochia on root pieces on filter paper and agar plates.

Rhizoctonia solani was rather common in 1978 and 1980. The fungus is mentioned as a root pathogen on strawberry (Montgomerie 1970). It has also been found in Finnish

strawberry roots (Ylimäki 1970), as have *Phoma* species, which were also present in this study. Unidentified *Phoma* species were found mostly in 1980.

Armillaria mellea, the honey fungus, was found each year in rotted or nearly rotted taproots. Such roots were soft, easily broken, light to dark brown, and with cavities filled with white mycelia in rotted tissue (Fig. 3 F). The strawberry plants with Armillaria root rot showed wilting and were often a reddish colour. The fungus was not common in plantations, but was found sporadically. A. mellea is also mentioned as a root and crown pathogen on strawberry by Wilhelm (1961). The fungus formed mycelia on root pieces and sometimes also rhizomorphs on agar plates (Fig. 3 G).

The Coniothyrium species C. fuckelii and C. fragariae are mentioned as pathogens on strawberry crowns (Jarvis and Hargreaves 1972). C. fuckelii was rather common in 1979 and 1980 in this study. C. fragariae was rare and present only in rotted or nearly rotted taproots.

Gnomonia fragariae was found sporadically in this material. The fungus was present in old, often weak plants showing a reddish colour in the older leaves. It formed perithecia on taproot pieces from plants demonstrating root rot and crown necrosis (Fig. 3 A, E). In autumn the fungus was found on petioles, causing black lesions and forming numerous black, globose perithecia with long necks (Fig. 3 B, C). The fungus can cause lesions and necrosis on leaves and petioles and can attack unripened fruits (Bolton 1954). G. fragarie is considered an important pathogen on strawberry fruits in central Europe (Seemüller 1969).

Verticillium spp. were rare in the material. Diplocarpon earliana causes leaf scorch on strawberry leaves, and was found sporulating on taproot surfaces, too.

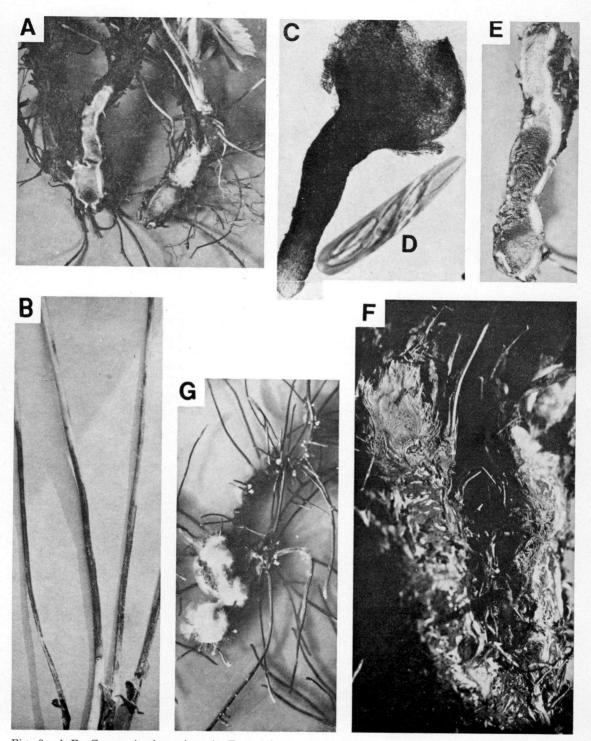


Fig. 3. A-E: Gnomonia fragariae. A, E: causing root rot in strawberry taproots. B: lesions and necrosis on petioles. C, D: perithecium and ascus. F-G: Armillaria mellea. F: root rot with white mycelia in rotted tissue. G: rhizomorphs on PDA-plate. Material: A, C, D: EH, Vanaja 10.10.1980, B, E, F: V, Vahto. B, E: 10.10.1980. F: 22.9.1980. G: St, Kokemäki 10.10.1979. C:  $\times$  100, D:  $\times$  500, G:  $\times$  1.

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#### SELOSTUS

#### Mansikan juurilaho

#### PÄIVI PARIKKA

#### Maatalouden tutkimuskeskus

Mansikan juurilahon aiheuttajien selvittämiseksi kerättiin vuosina 1978—1980 486 näytettä mansik-kaviljelmiltä, etupäässä Pohjois-Savosta ja Varsinais-Suomesta. Tutkittavaksi valittiin yleensä heikkokuntoisia kasveja vanhemmista kasvustoista. Kasvien juurakoista tarkastettiin vioitukset sekä määritettiin sienistö kosteakammio- ja agarkasvatuksista.

Juurilahoa aiheuttavista sienistä yleisimpiä olivat *Fusarium*-lajit, joita löytyi 62 %:ssa näytteistä. Määritetyistä kahdeksasta lajista yleisin oli

F. avenaceum. Cylindrocarpon-lajeja, pääasiassa C. destructansia oli 32 %:ssa näytteistä. Myös Botrytis cinerea oli yleinen ja sitä esiintyi 26 %:ssa juurakoista. Rhizoctonia solani, Hainesia lythri ja Coniothyrium fuckelii olivat hieman harvinaisempia. Juurakoiden nopeana lahottajana oli paikoin mesisieni Armillaria mellea. Gnomonia fragariae aiheutti mansikan juurakoissa lahoa ja lehtiruodeissa laikkuja. Sientä löytyi eri puolilta Suomea.

#### Seria PHYTOPATHOLOGIA N. 86 — Sarja KASVITAUDIT n:o 86

## VARIATION FOR SPECIFIC VIRULENCE IN THE FINNISH BREMIA LACTUCAE POPULATION

#### KIRSTI OSARA and IAN R. CRUTE

Osara, K. & Crute, I. R. 1981. Variation for specific virulence in the Finnish Bremia lactucae population. Ann. Agric. Fenn. 20: 198—209. (Agric. Res. Centre, Inst. Pl. Path., SF-01300 Vantaa 30, Finland).

Lettuce downy mildew (Bremia lactucae Regel) was found in Finland in the province of Uusimaa between 1972 and 1976 at 13 farms in 5 communes. The disease occurred on protected lettuce crops during September and October and in field crops during September. From eleven collections of the fungus, six different virulence phenotypes were recorded (SF1—6). Details of five of these phenotypes have not previously been reported. Virulence factors v1 to 11 were located in the Finnish pathogen population at varying frequencies. The most complex phenotypes (SF3 and SF6) carried 8 and 10 v-factors respectively while SF5 carried only two v-factors. Factor v2 was universally present in all isolates which probably reflects the common commercial usage of cultivars carrying R2. In contrast to other areas of Europe, v1 was not universally present in isolates. Complete resistance to all isolates could be contributed by R11 together with either R4, 6 or 10.

The specific resistance of 126 lettuce cultivars to the six distinct pathogen phenotypes was tested and there were a few anomalous reactions which could not be accommodated solely by the R-factor combinations previously postulated for certain cultivars. It is suggested that this is due to the occurrence of R1, previously undetected in these cultivars.

There was a significant difference between cultivars in the field susceptibility of adult plants grown in a Polythene greenhouse following inoculation by the multivirulent isolate SF6.

Index words: *Bremia lactucae*, downy mildew, lettuce, *Lactuca sativa*, physiologic races, resistance factor, virulence phenotype, cultivars, adult plant resistance, seedling resistance, disease distribution.

#### INTRODUCTION

Lettuce downy mildew (*Bremia lactucae* Regel) was first found in Finland in 1970. Damage leading to complete crop loss became common and the economic losses to growers

were considerable. The purpose of this study was to obtain information about the distribution of the disease in Finland, to examine the variation for specific virulence in the region and to locate those cultivars most likely to prove resistant.

Breeding for resistance to downy mildew has been an objective of lettuce breeders for more than 50 years. However, concentration on the utilisation of race specific major genes has resulted in only transient control of the disease. A gene-for-gene type of relationship exists between the host and pathogen with eleven resistance factors (R-factors) in lettuce cultivars matched by eleven comparable virulence factors (v-factors) in pathogen isolates (Crute and Johnson 1976, Johnson

et al. 1977, 1978). To provide information on those cultivars most likely to prove resistant in a locality it is necessary to determine the frequency of occurrences of particular virulence factor combinations in the pathogen population (Dixon and Wright 1978, Wellying and Crute 1978, Lebeda 1981).

There is little information of the relative field response to infection of commercial cultivars once they are attacked by *B. lactucae*. A further objective of this study was to investigate the variation between cultivars in this respect.

#### MATERIAL AND METHODS

Information about the distribution and virulence characteristics of *B. lactucae* in Finland was collected between 1972 and 1976 by investigating 65 lettuce samples obtained from 39 growers.

Most of the samples were from the provinces of Uusimaa (39 samples), Kymenlaakso (11 samples) and Varsinais-Suomi (6 samples).

B. lactucae was isolated from infected samples between 1974 and 1976 and the isolates were tested between 1975 and 1980. The fungus was isolated by washing spores from diseased leaves using distilled water. Leaf discs or seedlings of the same cultivar from which the isolate was taken or cv. Hilde were inoculated with the resulting spore suspension.

Isolates could be stored deep-frozen at —20°C for 6 months and would remain viable. Single spore cultures were produced by inoculating leaf discs of the cultivar from which the isolate was taken with one spore transferred on a piece of agar. Initially, tests were made using a set of ten differential cultivars (Hilde, Blondine, Noran, Ancora, Brioso, Portato, Kordaat, Solito, Calmar, Hilde x Lactuca serriola). Ten leaf discs,

1,7 cm diameter from each cultivar were inoculated using a small pipette (Tjallingii and Rodenburg 1969). The test was repeated using suspensions made from every susceptible cultivar separately to obtain information about the stability of the race and homogenity of the isolate.

A larger set of cultivars was used to ensure that the presence or absence of virulence to all R-factors 1—11 was determined (Table 1). For these tests, and for tests in which more than 100 cultivars were inoculated with six distinct pathogen virulence phenotypes seedlings at the cotyledon stage were used. The seedlings were grown for 7 days in seed trays in peat covered with sterilised sand first at a temperature of +20°C and after germination at a temperature of +17°C with a 16 h photoperiod and a light intensity of 4000 lx. Three replicates of 10 cotyledon pairs per cultivar were placed on their abaxial surface in 9 cm glass Petri dishes on two filterpapers moistened with distilled water.

The spore suspension was prepared by shaking infected seedlings or leaf discs in distilled water and washing three times by centrifugation (Dickinson and Crute 1974). There were  $1.5 \times 10^5$  spores/ml. The sus-

pension, 1 ml/Petri dish, was sprayed on to seedlings through the nozzle of a seed treater (Vanhanen 1977) at a pressure of 49 kPa. The Petri dishes were placed in a seed tray and covered with a transparent 0,5 cm thick plastic plate. The leaf discs and seedlings were incubated at a temperature of  $\pm 15 \pm 3$ °C under warm fluorescent tubes at a light intensity of 3000 lx with a 16 h photoperiod.

Records were made 7, 10 and 14 days after inoculation. Cultivars were categorised as complete resistant (recorded —) when no sporulation occurred, completely susceptible (recorded +) when sporulation was profuse on most seedlings, or incompletely resistant (recorded (—) (Crute and Norwood 1978) when sporulation was sparse. At the same time intervals records were made of the percentage of leaf area (60 cotyledons/cv.) covered with sporophores (Dixon and Doodson 1971).

In 1977 the severity of infection of 26 cultivars by isolate SF6 was studied under

field conditions. Cultivars were sown on 27, 7, in Paperpot Vh 505 1/2 in sterile peat and planted on either 9.8. or 10.8. in a Polythene house in peat at a spacing of  $20 \text{ cm} \times 20 \text{ cm}$ . Plots were arranged in randomised blocks and between every 5 cultivars there was a row of cv. Hilde. In every plot there were 10 plants with one plant of Hilde on both ends of the row. At the 3-4 adult leaf stage the plants were inoculated on 22.8. at 19.00 hours when the temperature was  $+ 19^{\circ}$ C using an AZO-propan sprayer. There were  $8.5 \times 10^4$  spores/ml in the inoculum which was applied at a rate of 2,5 ml/plant. During incubation the minimum night temperature was +2 - +6°C and the maximum day temperature + 19 - + 35°C. Half of the experiment was assessed 15 days after inoculation and the remainder after 19 days using the adult plant key categories: 0 (healthy), 5, 10. 25, 50, 75,  $100 \, \%$  of the leaf area affected (Dixon and Doodson 1971).

#### RESULTS

#### Disease occurrence and distribution

B. lactucae was found on 13 farms. These were situated in the province of Uusimaa in 5 communes: Espoo (5 farms), Kirkkonummi (4 farms), Nummi (2 farms) Helsinki (1 farm) and Pohja (1 farm) (Fig. 1). 25 lettuce samples were affected by B. lactucae (38,5% of the total number of samples received). These came from the following communes: Espoo (16 samples) Kirkkonummi (5 samples), Nummi (2 samples), Helsinki (one sample) and Pohja (one sample).

The fungus was detected in most years and on several cultivars at the same farm. A total of nine cultivars was affected. In greenhouses during September and October these were: Amanda, Attraction, Larganda, Noran, Ostinata, Plenox and Solito and outdoors during September the disease was found on Great Lakes and Market Favorite.

The virulence phenotypes SF1—6 were distributed as follows: in Espoo SF1 on cv. Larganda, Ostinata and Solito, SF3 on cv. Solito and SF5 on cv. Noran, in Helsinki SF4 on cv. Market Favorite, in Kirkkonummi SF1 on cv. Ostinata and SF2, SF4 and SF6 on cv. Noran, in Nummi SF6 on cv. Noran and in Pohia SF1 on cv. Great Lakes.

#### Virulence phenotypes and the identification of R1 in lettuce cultivars

From eleven collections of *B. lactucae* six different virulence phenotypes (SF1—6) were recorded (Table 1 and 2). Only SF1 has been

Table 1. Reactions of differential lettuce cultivars to Bremia lactucae isolates SF1-6.

Cultivar	Postulated R-factors	SF1	SF2	SF3	SF4	SF5	SF6
Hilde Blondine Cristallo. Dandie Valmaine Sabine Mesa 659 Valverde Bourguignonne Tornado	0 1 2 (+ 1) b 3 5 6 7 8 9	+ + + + + (_) + + (_)	+ + + (-) - + +	+ + + + ( <u>-</u> ) + ( <u>-</u> )	+ a () + + + + +	+ 	+++++++++
Hilde × L. serriola Noran Ancora Brioso Portato Larganda Parmanta Edgar Kordaat Solito Diana Bremex Calmar Ardente Avondefiance Avoncrisp	11 $2+4$ $2+4$ $2+7$ $(+1)^{b}$ $2+7$ $(+1)^{b}$ $2+7$ $2+11$ $2+3+7$ $3+4$ $3+7$		+ + + + - - - + +	+ + + + + + + + + + + + + + + + + + + +		+ a	+++++++++++++++++++++++++++++++++++++++

a = Anomalous reactions not consistent with previously postulated R-factor complement of cultivars
 b = See text for explanation

previously reported in published work (Wellving and Crute 1978, Norwood and Crute 1980). From one collection two different virulence phenotypes were separated (SF1 and SF3).

Table 1 gives the results of tests on twenty-six differential cultivars which cover the range of postulated R-factor combinations available in cultivated lettuce types. The majority of reactions were entirely in accordance with the predictions of the genefor-gene model postulated by Crute and Johnson (1976) or its modifications (Johnson et al. 1977, 1978, Norwood and Crute 1980).

There were, however, a few anomalous results which indicated the need to modify

the postulated R-factor complements of certain differential cultivars by suggesting the presence of R1 which had been undetected previously.

The cultivar Blondine, which carries resistance factor R1 only, was resistant to isolates SF4 and SF5 but susceptible to SF1, SF2, SF3 and SF6. Both SF4 and SF5 carried v2 and v4 recognised by virulence on cv. Noran (R2  $\pm$  4) and other cultivars with the same R-factors (Table 4). Nevertheless, some cultivars which would be expected to prove susceptible to either or both of SF4 and SF5 were shown to be resistant i.e. Cristallo (thought to be R2 only), Ancora (thought to be R2  $\pm$  4), Portato (thought to be R2  $\pm$  7),

<sup>+ =</sup> Susceptible

<sup>— =</sup> Resistant

<sup>(—) =</sup> Incomplete resistance with sparse sporulation

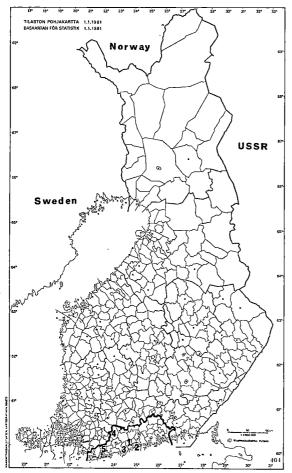


Fig. 1. The distribution of *Bremia lactucae* in Finland 1972—76. 1. Espoo, 2. Helsinki, 3. Kirkkonummi, 4. Nummi, 5. Pohja.

Bremex (thought to be R4+7) and Ardente (thought to be R4+6+7). Postulating the presence of R1 in each of these cultivars and others reacting like them, would explain the apparent anomalies. Cvs. Bremex and Ardente may carry R1 and not R4 but this is not proved (see reaction to SF1). It is still possible, in addition, that any cultivar carrying R3 could also be carrying undetected R1 since there are no isolates available which carry v3 but which are avirulent on cv. Blondine (i.e. without v1). Alternatively, if factor R1 is not present in the above cultivars,

Table 2. Postulated virulence phenotypes of Bremia lactucae isolates SF1-6.

Race nomen- clature				Vir	uler	ice :	phe	noty	pe			No. of times re- corded *
SF1	1	2	3	×	5	×	7	8	×	10	X	5
SF2	1	2	X	4	X	6	7	×	X	X	X	1
SF3	1	2	3	$\times$	5	X	7	8	9	X	11	$\bar{1}$
SF4	$\times$	2	$\times$	4	5	6	7	8	9	10	X	<b>2</b>
SF5	×	2	X	4	X	$\times$	X	X	X	X	X	1
SF6	1	2	3	4	5	6	7	8	9	10	$\times$	2
										To	tal	12

<sup>\*</sup> The presence or absence of v5, v6, v9, v10 was tested for only one isolate of each race.

then some new and previously undetected R-factor must be implicated.

Table 2 gives the postulated virulence phenotypes SF1—6 and the number of times each was recorded. SF6 is the same phenotype as the isolates 74/T (also referred to as Tv) (Crute and Davis 1976), S4 (Wellving and Crute 1978) and  $N_6$  (Lebeda 1980). SF5 is an isolate carrying only two virulence factors and is the most simple isolate to have been reported.

The virulence phenotype of SF1 differs from that previously published by Wellving and Crute (1978), due to the re-interpretation of results related to the presence or absence of R4 in some genotypes as explained by Norwood and Crute (1980), and also to an error in those publications with respect to the presence of v1, v9 and v11.

#### v — factor frequencies

The number of isolates studied (12) is too few to draw firm conclusions about the significance of the observed v-factor frequencies (Table 3). However, there are some observations of interest. The universal presence of v2 in the isolates sampled contrasts with the situation reported for nearby Sweden (Well-

Table 3. Observed frequencies of v-factors 1 to 11 in the Finnish *Bremia lactucae* population. Calculated from a sample of 12 isolates.

v-factors	Frequency
1	0,75
$\overline{\overset{-}{2}}$	1,00
3	0,67
4	0,50
5	0,83
6	0.42
. 7	0,92
8	0,83
9	0,50
10	0,75
11	0,08

ving and Crute 1978) where this v-factor was infrequently encountered. In Czechoslovakia v2 was also reported to be universally present in the pathogen population (Lebeda 1979, 1981).

With the exception of v11, most other vfactors occurred at a relatively high frequency in the pathogen population. In common with other areas of Europe, cultivars carrying R11 would prove to be resistant more often than others. When the observed and expected frequencies of virulence factor combinations were compared following the methods of phenotype analysis (Wolfe et al. 1976) it became apparent that v-factors 5, 8 and 10 were more frequently combined than would be expected from their individual observed frequencies. This phenomenon has been commonly encountered in surveys of B. lactucae populations elsewhere (Wellving and Crute 1978, Dixon and Wright 1978, Crute and Dixon 1978, Lebeda 1981). Since in this survey no isolates came from cultivars carrying R5, 8 and 10 and such cultivars are not grown in Finland, it suggests that there are advantages to the pathogen in assembling these v-factor combinations additional to the specific virulence function.

#### Seedling reactions of cultivars to SF1-6

Table 4 shows the reaction of all the lettuce cultivars tested to isolates SF1—6. Cultivars have been grouped according to their postulated R-gene complements from this study and others. It can be seen that SF1—6 do not discriminate between cultivars with R3, R3 + 7, R2 + 3 + 7 and R3 + 7 + 8 which all give the same reaction array with these isolates. It is therefore not possible to determine from these reactions, the R-gene complement of several numbered lines listed at the end of Table 4.

There is some evidence from this data that SF2 may invoke an incomplete resistance reaction in cultivars resistant by virtue of R3. This is unusual since R3 normally conditions a complete response associated with very restricted microscopic fungal development.

The reaction pattern of cv. America is not explicable in terms of R1—11 and requires further examination. The only other anomalous reaction in the set of data in Table 4 concerns cv. Sabine (and possibly cv. Tires which may also carry R6) and SF1. Usually cv. Sabine gives an incomplete resistance response to those isolates lacking v6 such as SF1 characterised by sparse and retarded sporulation (see Table 1). In Table 5, Sabine is clearly reacting as fully susceptible. It is possible that there may be variation between seed batches which were different for the two sets of tests.

#### Adult plant reaction of cultivars to SF6

All cultivars susceptible to SF6 (v1—10) were attacked in the Polythene house. There were some differences in different parts of the house, but nevertheless there was a significant difference between cultivars in the degree of infection (Table 5).

Table 4. Seedling reaction of lettuce cultivars to Bremia lactucae isolates SF1-6.

Number	Cultivar	Postulated R-factor		SF1	SF2	SF3	SF4	SF5	SF6
	Hilde	0				,			
52	Amerikans Ploksallat WW	0 0		+	+	+	+	+	+
4	Aurelia Cl			+	+	+	+	+	+
6	= =	0		+	+	+	+	+	4
54	Bellona BS	0	- '	+	+	+	+	+	4
	Bergina VDB	0		+	+	+	+	+	+
106	Duna SP	0		+	+	+	+	+	+
.08	Grinlek (F 003) Sv	0		+	+	+	+	+	+
59	Helga NZ	0		+	+	+	+	+	+
61	Helresta BS	0		+	+	+	+	+	+
62	Hjerter Es Enkona P 68	0		+	+	+	+	+	+
63	Irma DP	0	`	+	+	+	+	+	÷
65	Lavina BS	0		+	. +	+	+	+	+
66	Magna VDB	.0		+	÷	+	+	÷	+
68	Marco DP	0		+	÷	÷	+	÷	÷
69	Market Favorite WW	0 -		+	÷	÷	+	4	+
71	Massa DP	0		÷	÷	+	÷	+	
72	Mathilde OE	Ô		+	+	+	+	+	+
73	Maturda VDB	Õ		+	+	+			+
75	Olof NZ	ő		7	7		+	+	+
31	Plenos OE	0		+	+	+	+	+	+
76	Prado RS	0		+	+	+	+	+	+
79	Resistent RS	=		+	+	+	+	+	+
82		0		+	+	+	+	+	+
85 ·	Rigoletto RS	0		+	+	+	+	+	+
	Sanno DP	0		+	+	+	+	+	+
86	Saskia RZ	0		+	+	+	+	+	+
87	Silvester NZ	0		+	+	+	+	+	+
88	Steran VDB	0		+	+	+	+	+	+
89	Suzan SG	0		÷	÷	. +	÷	÷	+
91	Vigar DP	` 0		÷	+ .	÷	÷	÷	+
92	Viresta BS	Ō ,		÷	÷	+ .	+	<u> </u>	÷
93	Virilde (843—71) NZ	Ô		÷			1.	- -	7
94	Viruzan (IIo) SG	0		-	+	+.	+	+	+
95	Zommerdiamant BS			+	+	+	+	+	+
99	No 650 OE	0		+	+	+	+	+	+
00	110 030 CE	0		+	+	+	+	+	+
	Blondine	1		+ '	+	.+			+-
02	Dandie NSDO								
28 .	Ostinata VDB	3		+	()	+	(—)		+
34	Sabine EZ	3		+		+		_	+
)4		6		+	+	(—)	+	()	+
	Tires (F 006) Sv	6 ?		+	+	<del></del>	+	(—)	+
38	Tornado BS	10		+			+	_	+
	Hilde  imes L, $serriola$	4.4					•		
51	Ameku SG	11				+	_	_	<u>-</u>
56		11		_	_	+	_	_	_
	Capitan SG	11		_		+	(—)	()	
7	Bremex SG	1+7		+	+	+	·		+
	Noran			•		•			
3		2+4			+	_	+	+	+
-	Arosa-Let (10087) DP	2+4	5.		+	_	+	+	+
.2	Deci-Minor RZ	2+4			++++	_	++	+	+
	Deciso RZ	2+4			+		+	+	+
7	Knap SG	2+4		(—)	+	_	+	+ + +	+
	Lucia Cl	2+4		_	+		÷		+ + + +
	Maikönig WW	2+4			÷	_	+	+ + +	<u> </u>
2	Miranda DP	2 + 4			÷		+	1	ユ
	Plus (Amanda Pl.) DP	2 + 4			<u> </u>	_			+
	Selma BS	2+4		.—	1		+	+	+
_	Wintosa VDB	$\frac{2+4}{2+4}$		_	+ + + +		+	+	+ + + +
3	495/74 S BS				+	_	+	+	+
		2+4		(—)		_	+	+	+
	No 117 DP	2+4			+		+	+	+
5	No 1600 DP	2 + 4		_	+	_	+	+	+
	Etam Cl	2+4		NT	NT		+	+	NT
	O1	A T +		DI I	INCT:				70,777

<sup>+,</sup> susceptible; —, resistant; (—), incomplete resistance with sparse sporulation; NT, not tested. Cultivars tested with single spore isolates: SF1, 1—110; SF2, 1—110; SF3, 51—110; SF4, 51—110.

	<u> </u>							
Number	Cultivar	Postulated R-factor	SF1	SF2	SF3	SF4	SF5	SF6
50 21 33 35 77	Larganda RZ Mandela (544) DP Ravel (50) RZ Rossini (3482) RZ Type 50 OE	$egin{array}{c} 2+7 \\ 2+7 \\ 2+7 \\ 2+7 \\ 2+7 \\ 2+7 \end{array}$	+ + + +	+ + + + +	+ + + +	++++		+ + + +
20 30	Mandaat (A342) SG Parmanta (A308) SG	$2+11 \\ 2+11$	_	_	+ +	_	_	_
16 18	Kordaat Kloek SG Kwiek SG	$   \begin{array}{r}     3+4 \\     3+4 \\     3+4   \end{array} $	( <u>-</u> )		<u>-</u>	1 -	=	+ + +
107 8 55 9 15 64 70 23 26 32 80 81	Solito Bizet (2598) RZ Brevier SG Brezan (Cal) SG Cynthia (27) EZ Helia (71) EZ Kares RZ Mariska (Bora) SG Mistra (1596) OE Novaran OE Plevanos RZ Reskia RZ Respons OE	3+7 3+7 3+7 3+7 3+7 3+7 3+7 3+7 3+7 3+7	+++++++++++++++++++++++++++++++++++++++		++++++++++++			++++++++++++
103	Avondefiance NSDO	6+8	(—)		(—)	. +		+
53 24	Calmar Appia Cl Mania Cl	7+8 7+8 7+8	+ + +		+ + +	+ + +	_ _ _	, <del>+</del> , <del>+</del>
2 10	Ancora Amplus 75 VDB Dalida DP	1+2+4 $1+2+4$ $1+2+4$	  ( <del>(</del> )	+ + +	_  ()	=	=	+ + +
27 34 41 78	Portato Brioso Orlando (173) DP Renate (70) EZ Winos (3000) DP Type 70 OE	$egin{array}{c} 1+2+7 \\ 1+2+7 \\ 1+2+7 \\ 1+2+7 \\ 1+2+7 \\ 1+2+7 \end{array}$	+ + + + + +	+ + + + +	+++++++++++++++++++++++++++++++++++++++		1 1 1 1	+ + + + + +
13 46 40 29	Edgar RS Orba (B67) SG Wenda SG Pallas DP	2+3+7 $2+3+7$ $2+3+7$ $2+3+7$	+, + +	_ / _ _				+ + + +
47 58 90 25	Corelli (9786) RZ Diana (965) Tozer Verpia Cl Nordia (61) EZ	3+7+8 ? $3+7+8$ $3+7+8$ $3+7+8$ ?	+ + + +	_ 	+ + +		- - -	+ + +
110 96 97 5 98 100 49 48	B 3 P No 75117 DP No 75136 DP No 3 OE No 59 OE No 1447 OE No 338 EZ No 444 EZ		+ + + + + + +		++++++++++++	   NT		+++++++++++++++++++++++++++++++++++++++
57 83 74 60 101 36	Type 27 OE Type 51 OE Type 61 OE Type 71 OE Resto (F 005) Sv Salina (63) DP		+ + + + + + + + + + + + + + + + + + + +		.+++++			++++
39 109	Vera (Vox) SG America SG	. ?	· +	+	+	+	+	+
100		•	<u>'</u>				Т	T

Source of the varieties: BS, Bruisma Seed; Cl, L. Clause; DP, D. v.d. Ploeg; EZ, De Enkhuizer Zaadhandel; NSDO, National Seed Development Organisation; NZ, Nunhems Zaden; OE, J.E. Ohlsens Enke; P, C. W. Pannevis; RS, Royal Sluis; RZ, Rijk Zwaan; SG, Sluis & Groot; SP, Carl Sperling & Co; Sv, Sveriges Utsådesförening; VDB, v.d. Berg; WW, W. Weibull Ab.

Table 5. The severity of infection of seedlings and adult plants by SF6 isolate.

Cultivar	Postulated R-factors	Leaf are	a affected %
		Seedlings	Adult plants
Tornado BS	10	44	84
Ostinata VDB	3	40	83
Dandie NSDO	3	44	82
Plevanos RZ	3+7	35	79
Nordia (61) EZ	3+7+8?	38	77
Renate (70) EZ	1+2+7	40	71
Edgar RS	2+3+7	16	63
Brevier SG	3+7	30	61
Kloek SG	3+4	39	58
Kwiek SG	3 + 4	36	58
Ravel (50) RZ	2 + 7	30	58
Amplus 75 VDB	1+2+4	43	55
Γires Sv	6 ?	46	53
Avondefiance NSDO	6 +- 8	36	52
Deci-Minor RZ	2+4	35	52
197/74 S BS	2+4	20	50
75 11 7 DP	?	44	48
Resto Sv	?	35	45
Hilde NZ	0		43
Selma BS	2+4	19	41
Reskia RZ	$^{3+7}$	48	40
Mania Cl	7+8	30	39
Capitan SG	11	4	4
Ameku SG	11	0	Ô
Mandaat SG	2+11	0	ő
Parmanta SG	2+11	0	ő
			F 15,42 ***
			LSD % 12,9

In seedling tests, cv. Capitan only showed necrotic flecking, but in this experiment some sparse sporulation was observed.

All R-factors except R5 and R9 were present in the cultivars inoculated with SF6 and there was not a significant correlation between the percentage of affected leaf area and the R-factors present in the cultivar. However, some cultivars with the same R-factors had almost the same percentage of damage, e.g. R3 Ostinata 83 % and Dandie

82 %, R3 + 4 Kloek 58 % and Kwiek 58 %, R2 + 4 Deci-Minor 52 %, 494/74 S 50 % and Selma 41 %.

The correlation between the percentage of leaf area covered with sporophores on cotyledons 10 days after inoculation and the percentage leaf area affected in this experiment was highly significant (r = 0.80\*\*\*) but when cultivars with R11 and R2 + 11 were not considered there was no correlation (r = 0.25).

#### DISCUSSION

Lettuce downy mildew was found only in the province of Uusimaa between 1972 and 1976. In this district the commercial farms are small and lettuce is grown every year. The sale of both home produced and imported lettuce occurs at the same market and it is possible that the disease has been spread from infected imported lettuce.

The results showed that virulence factors v1 to 11 are all represented in the Finnish B. lactucae population but there were differences in their frequency. The fact that factor v2 was present in every isolate is probably a result of R2 being present in cultivars, e.g. Amanda, Larganda and Noran, commonly grown at the time the population was sampled. By comparison, v1, 4 and 6 were all present at lower frequencies than have been recorded in some other surveys (Dixon and Wright 1978, Wellving and Crute 1978, Crute and Dixon 1978).

The resistance factor R1 primarily characterised by the resistance of cv. Blondine is rarely recorded as effective against B. In other words, v1, the lactucae isolates. virulence factor matching R1 and rendering it ineffective is very frequent and widely distributed throughout the pathogen population. In the UK, v1 was shown to be present at a frequency of 1,00 (i.e. in a survey carried out between 1976 and 1978 all isolates tested were pathogenic on cv. Blondine) (Crute and The best documented isolate Dixon 1978). which Blondine is resistant to is NL3 which has the virulence phenotype v: 5, 6, 7, 8, 10 (Crute and Johnson 1976, Blok and van der Schaaf-van Waadenoyen Kernekamp 1977). At NVRS, a UK isolate (IM 43) which has the same virulence phenotype has been studied. Since these two isolates also lack v2, 3 and 4, any cultivar which is resistant to these isolates by virtue of R2, 3 or 4 or any combination of these factors (proved following experiments with other isolates) may also carry R1 undetected (see also footnote e, Table 4 in Crute and Johnson 1976). reaction patterns of the isolates of B. lactucae

from Finland suggest that this may have occurred. Since R1 is rarely effective against B. lactucae in Europe at least, these findings have little practical relevance. They demonstrate, however, that the presence of R1 previously undetected in certain commercial lettuce cultivars, may have resulted in selection for v1 and hence explain more readily its frequent occurrence in the population.

In common with other regions of Europe where studies have been conducted, factor v11 was found to be present at the lowest frequency. The relatively new factor, R11 particularly combined with other R-factors, R4, 6 or 10, is therefore most likely to provide protection from the disease in Finland.

Variation for the field susceptibility of lettuce cultivars was demonstrated when a range of cultivars was inoculated with the multivirulent isolate SF6. However, when cultivars resistant by virtue of R11 were eliminated from the analysis there was no correlation between disease severity on seedlings and that on adult plants. There have been comparatively few studies on the relative field susceptibility of lettuce cultivars, but those reported confirm that variation exists which could possibly be exploited both by plant breeders and growers (Dixon et al. 1973, Crute and Norwood 1981).

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#### SELOSTUS

## Salaatin lehtihomeen aiheuttajan Bremia lactucae saastutuskyvyn vaihtelusta

#### KIRSTI OSARA ja IAN R. CRUTE

Maatalouden tutkimuskeskus ja National Vegetable Research Station

Salaatin lehtihomeen (*Bremia lactucae* Regel) levinneisyyttä selvitettiin vuosina 1972—1976. Salaattinäytteitä saatiin eri puolilta maata, pääosan ollessa Etelä-Suomesta kerättyjä. Tautia tavattiin ainoastaan Uudenmaan läänin alueella 13 viljelmällä. Tutkituista näytteistä oli 38,5 % taudinaiheuttajan saastuttamia. Tautia esiintyi avomaalla syyskuussa ja kasvihuoneissa syys-lokakuussa.

Testikasvien avulla määritettiin sienieristyksistä kuusi virulenssifenotyyppiä SF1-6, joista ainoastaan SF1 on aikaisemmin selostettu kirjallisuudessa.

Virulenssitekijöiden, v1—11 lukumäärät vaihtelivat eri fenotyypeissä 2—10 tekijään. Tietyt tekijät esiintyivät toisia lukuisammin. Poiketen aikaisemmin Euroopassa julkaistuista tiedoista,

tekijä v1 puuttui osasta isolaatteja, kun taas tekijä v2 esiintyi kaikissa isolaateissa.

Täydellinen taudinkestävyys on saavutettavissa viljelemällä lajikkeita, joilla resistenssitekijä R11 on yhdessä R4, 6 tai 10 kanssa.

Sirkkalehtiasteella olevilla taimilla määritettiin 126 salaattilajikkeen taudinkestävyys erikseen jokaista patogeenista fenotyyppiä vastaan. Testauksessa ilmeni, että eräät lajikkeet sisältävät ennen oletettujen resistenssitekijöiden ohella myös tekijän R1.

Muovihuoneessa kasvatettujen salaattilajikkeiden kenttäkestävyydessä oli merkitsevä ero fenotyyppiä SF6 vastaan, jossa on virulenssi-tekijät v1—10.

#### Seria PHYTOPATHOLOGIA N. 87 — Sarja KASVITAUDIT n:o 87

# RESEARCH NOTE THE FIRST REPORT OF LETTUCE MOSAIC VIRUS IN FINLAND

#### MARJA-LEENA LAHDENPERÄ

Lahdenperä, M-L. 1981. The first report of lettuce mosaic virus in Finland. Ann. Agric. Fenn. 20: 210—213. (Agric. Res. Centre, Inst. Pl. Path., SF-01300 Vantaa 30, Finland).

A virus disease occurring in some lettuce samples sent by growers in Finland proved to be lettuce mosaic. The virus was identified on the basis of the symptoms it induced in various test plants and by determining its thermal inactivation point and measuring the length of the virus particles in electron microscope.

Index word: Lettuce mosaic virus.

Samples of diseased lettuce grown under glass, cultivar unknown, were sent to the Institute of Plant Pathology in 1980 by a farmer in Varkaus in eastern Finland. Lettuces with somewhat similar symptoms have been sent earlier, too, but the cause of the disease has not been diagnosed. The leaves of the plants were much smaller and thicker than usual. The veins, especially the mid rib, were big compared with the size of the leaf blade, which moreover was curly. The disease was suspected to be caused by lettuce mosaic virus (LMV). The mosaic in young plants is easily recognized, but later the symptoms become indistinct and highly variable (Grogan 1980). Tests were consequently performed to find out the real cause of the disease.

#### Virus symptoms in lettuce plants

It was necessary to make some inoculations to different lettuce cultivars, because the symptoms of lettuce mosaic vary greatly. Five cultivars of lettuce were used in infection tests. They were butterhead lettuces »America», »Nya Hilde» and »Ostinata», a crisphead lettuce »Grinlek» and a leaf lettuce »Salad Bowl».

Sap transmission using phosphate buffer, pH 7,2, and carborundum powder as an abrasive was made from the diseased lettuce plants to cultivars mentioned above. A pre-inoculation dark treatment was used to promote the appearance of the symptoms.

The virus discussed was easily transmitted by sap. It produced somewhat similar symp-

toms in all lettuce cultivars used in this test. The sap inoculation usually caused systemic infection in 6-10 days, but sometimes light brown small necrotic spots appeared on the inoculated leaves as a first sign of the infection. The systemic symptoms consisted mainly of deformed leaves with mottling, curling and sometimes slight vein clearing. The surface of the leaf blades was rough and their edges turned downwards, so that the appearance resembled that of a spoon. Severely infected plants were dwarfed and slow in growth (Fig. 1). They failed to »heart», the leaves remaining small and rosetted. Later the plants turned yellow. The yield of these plants will probably remain very small.

Although the symptoms are highly variable, they agree closely with the notes in literature (cf. Tomlinson 1970). All lettuce cultivars used in this test were infected. Also, according to Grogan (1980), nearly all cultivars are susceptible to lettuce mosaic. In spite of this there was a difference in the severity of the symptoms among the lettuce cultivars in this experiments. Cv. Grinlek, which is a cripshead lettuce, appeared to be more tolerant than others, for in other lettuce cultivars all the 8 test plants were infected while in cv. Grinlek only one of the 8 plants showed symptoms.

#### Other host plants and symptoms of LMV

Besides lettuce, 9 plant species were inoculated mechanically. 4 of them showed symptoms typical of lettuce mosaic.

Chenopodium amaranticolor Coste et Reyn. produced small chlorotic local lesions with necrotic centre in 6—7 days. The systemic reaction appeared as yellow mottling combined with deformation and twisting of the upper leaves.

In C. quinoa Willd. the symptoms appeared as numerous necrotic local lesions 1 mm in



Fig. 1. Systemic crinkling, mosaic and stunted growth in lettuce cv. Ostinata infected with LMV 7 weeks after inoculation. Uninoculated control on the right. Photo R. Ylimäki.

diameter in 6—7 days, and systemic symptoms after 2 or 3 weeks showed yellow mottle and twisted and stunted apical leaves.

Nicotiana tabacum L. Samsun produced yellow faint lesions and seminecrotic spots in 6 days on the inoculated leaves.

In *Pisum sativum* L. cv. Kiri systemic yellow flecks appeared 2 weeks after the inoculation.

The symptoms described above were similar to those mentioned in the literature (cf. Costa and Duffus 1958, Tomlinson 1970, Klinkowski 1977).

The virus did not infect the following plants: Cucumis sativus L. Butcher's OE Special, Cucurbita pepo L. cv. Vegetable Marrow, N. clevelandii Gray, N. glutinosa L. and Petunia hybrida Vilm. cv. Resisto Rosa.

# Soil transmission

Some tests were made to discover whether the virus causing the disease under consideration is soil transmissible.

The soil on which the diseased lettuce plants were grown was preserved and kept dry at room temperature. The soil sample was later mixed with steamed soil to increase its volume, so that new plants could be transplanted there. Small plants of lettuce cv. Ostinata in 2—4 leaf-stadium were transplanted to the soil, in which cv. America was sown, too, but none of the plants were infected. The results indicated that soil-borne viruses could not be involved.

#### Virus identification

The virus was mainly identified on the base of the transmissibility by sap and according to the symptoms in different test plants. This was confirmed by the thermal inactivation point (TIP). The sap for this test was pressed from infected lettuce plants. TIP was determined by heating lots of 2 ml for ten minutes at various temperatures and testing them for infectivity on *C. quinoa*. The test indicated that the virus was inactivated when the sap was heated at 55°C, but some infectivity was retained at 52°C. The result agrees well with the experiments of Klinkowski (1977) who

mentions that the TIP of LMV is  $54-56^{\circ}$ C, while Tomlinson (1970) states that it is  $55-60^{\circ}$ C.

Some preparations were made to inspect the virus particles in electron microscope. The specimens from the sap pressed from the experimentally infected lettuce plants were prepared with the dip method after Brandes (1957) or after the method of Lesemann (1972). The preparations showed flexuous filamentous virus particles about 770 nm in length, but the virus concentration was unfortunately so low that only a few particles were seen, and the measurements are consequently not very reliable. According to Tomlinson's (1970) measurements the length of LMV particles is approximately 750 nm.

The virus damaging the lettuces was identified as lettuce mosaic virus by its transmissibility by sap, its symptoms in different test plants and its thermal inactivation point and by measuring the length of the particles. This is the first recorded report of the occurrence of the lettuce mosaic virus in Finland.

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#### SELOSTUS

# Salaatin mosaiikkivirus todettu Suomessa

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# Maatalouden tutkimuskeskus

Kasvitautien tutkimuslaitokselle tutkittaviksi lähetettyjen salaatin taimien kasvu oli hidastunut, eivätkä ne olleet kerineet kunnolla, vaan kasvit muodostivat tiheän lehtiruusukkeen. Lehtilapa oli pienikokoinen, voimakkaasti kupruileva ja suonet, erityisesti keskisuoni, olivat paksuntuneita. Oireet viittasivat salaatin mosaiikkivirukseen. Tällaisia salaatin taimia on tavattu meillä aikaisemminkin, mutta taudinaiheuttaja on jäänyt selvittämättä.

Taudinaiheuttajaviruksen toteamiseksi suoritettiin kokeita eri ilmaisinkasveilla. Samoin selvitettiin viruksen siirtymistapa. Virusta ei saatu siir-

tymään maan kautta, mutta sen sijaan se siirtyi helposti mehussa. Infektiokokeet eri salaattilajikkeilla osoittivat, että viruksen aiheuttamat oireet vaihtelivat suuresti salaatin eri kehitysvaiheissa.

Virus määritettiin salaatin mosaiikkivirukseksi siirtymistapansa, ilmaisinkasveihin aiheuttamiensa oireiden, lämmönsietorajan sekä elektronimikroskopoinnin perusteella.

Nyt esitetty tiedonanto on ensimmäinen ilmoitus salaatin mosaiikkiviruksen esiintymisestä Suomessa.

## Seria PHYTOPATHOLOGIA N. 88 — Sarja KASVITAUDIT n:0 88

# VIRUS DISEASES IN CARNATION AND CHRYSANTHEMUM CUTTINGS IMPORTED INTO FINLAND

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During 1978—1979 a total of 93 samples of imported carnation cuttings. mainly from three cultivars, and 203 samples of imported chrysanthemum cuttings of four cultivars were tested by means of test plants, antisera and electron microscopy. 51,4 % of the carnation cuttings were infected. The carnation cuttings were most commonly infected by mottle and vein mottle virus, which occurred in 40,8 % of the tested carnation samples in 1979. Etched ring virus was quite common, too. It was found in 29,5 % and 28,6 % of samples in 1978 and 1979 respectively. Ringspot virus was detected in one sample only. Besides the samples mentioned above, 25 samples of farmers' stocks were tested, too. With one excention, the same viruses were found as in the imported cuttings. In electron microscope examinations, flexuous 1150-1350 nm long particles like those of carnation necrotic fleck virus were seen. The virus, very probably necrotic fleck virus, was transmitted by the aphid Myzus persicae Sulz. from carnations into other Dianthus species and to Silene armeria L.

There was some difference in the amount of virus infections among carnation cuttings of different origins. The cuttings produced in Teneriffe and in the Federal Republic of Germany were quite healthy, whereas cuttings from Swedish firms propagated in Portugal were highly virotic. 18,2% of the imported chrysanthemum cuttings were virus infected. They were infected mainly by the chrysanthemum B-virus while a couple of samples were infected by aspermy virus and stunt viroid. The same viruses occurred in 10 samples taken from farmers' cultivations.

Index words: Carnation viruses, mottle, vein mottle, etched ring, ringspot, necrotic fleck, chrysanthemum viruses, aspermy, virus B, stunt viroid, Silene armeria, Saponaria vaccaria, Dianthus chinensis, D. deltoides, D. plumarius L.

## INTRODUCTION

Carnation and chrysanthemum plants are have spread, through the increasing interhighly susceptible to viruses. These viruses national trade in cuttings, to every country where carnations and chrysanthemums are grown.

Viruses occurring in countries where cuttings are produced are especially important. Unfortunately, most reports concern the occurrence of carnation and chrysanthemum viruses in Central European countries and not in Mediterranean countries.

The most common carnation virus is mottle virus (Kassanis 1955, Brierley and Smith 1957, Zandvoort 1973), which occurs also in some Mediterranean countries (Poupet et al. 1970). Vein mottle virus has not been common in northern and western Europe (Åhman 1969, Hollings et al. 1977) but it is common in Mediterranean countries (Poupet and Marais 1973).

Etched ring virus has been found in many European countries (Hollings and Stone 1961, Kristensen 1964, Hakkaart 1968, Åhman 1969) and in Mediterranean region in Israel (Smookler and Loebenstein 1975).

Ringspot virus occurs nowadays only sporadically (Hollings and Stone 1965, Åhman 1969).

Some years ago a new carnation virus, carnation necrotic fleck virus, was found in Japan (Inouye and Mitsuhata 1973). Later it

was detected in Israel (Smookler and Loebenstein 1974), in Italy (Rana et al. 1977) and in the USA (Mayhew 1979). Carnation streak virus, which occurs in France, is very similar to necrotic fleck virus (Poupet et al. 1975).

The most common viruses in chrysanthemum are aspermy and B-viruses (Hollings and Stone 1971, 1972). Chrysanthemum stunt viroid is spread where ever chrysanthemums are grown (Dimock 1947, Diener and Lawson 1973).

No special attention has been paid earlier to virus diseases of carnations and chrysanthemums in Finland, but in recent years carnation and chrysanthemum growers in Finland have noticed an increasing incidence of virus diseases in their stocks in glasshouses. The carnations and chrysanthemums are not propagated in Finland, but the cuttings are imported. There was some difference of opinion between farmers and importers on the origin of the viruses. In order to find out which virus diseases occur in carnations and chrysanthemums, and whether they come with infected cuttings or infect the plants later in the glasshouses, carnation and chrysanthemum cuttings were tested in 1978— 1979.

# MATERIAL AND METHODS

Tests were made in glasshouse with screened openings. In winter the test plants were illuminated by mercuric vapour lamps. In winter the temperature varied from +18—23°C, in summer +20—35°C. Samples of imported cuttings were taken by the officials of the Plant Quarantine Inspection Service before the cuttings were handed over to the growers. A sample consisted of about 10 cuttings from which all the leaves were used for the sap inoculation. In total 93 samples were taken, mainly from the three cultivars Lena, Scania 3C and White Sim. In all 203 samples

from chrysanthemum cuttings, cvs. Dramatic, Fandango, Hurricane and White Marble, were taken for testing. Furthermore, 27 carnation samples and 10 chrysanthemum samples were taken from growers' crops. Then plants with symptoms indicative of possible virus infection were chosen for samples.

The virus infection was detected by symptoms in indicator plants inoculated by sap, grafting or vector (aphid, *M. persicae*), testing by antisera, or electron microscope inspection.

The sap transmission was done by sap ex-

tract pressed from fresh or deep-frozen leaves. Phosphate buffer 0,2 M pH 7,2 was added to the sap from carnations and 0,5% sodium sulfate with the same buffer was added to the sap from chrysanthemums. Carborundum was used as an abrasive.

In sap transmission tests the following plant species were regularly used for carnations:

Chenopodium quinoa Willd.

Dianthus barbatus L. cv. Diadem Saponaria vaccaria L. cv. Pink Beauty Silene armeria L.

The samples were often also tested by using C. amaranticolor Coste et Reyn.

D. caryophyllus L. cv. Joker

D. chinensis L.

D. deltoides L.

D. plumarius L.

Gomphrena globosa L.

The following plant species were used when chrysanthemums were tested:

C. amaranticolor

N. clevelandii Gray

N. glutinosa L.

N. tabacum L. Samsun

Petunia hybrida Villm. cv. Resisto Rosa

Some of the chrysanthemum cuttings from each sample were tested on the chrysanthemum plants cvs. Deep Ridge, Fanfare and Mistletoe. A test plant top was grafted to a rooted cutting.

The aphid *M. persicae* was used as a vector to isolate and differentiate some carnation viruses. The aphids were starved for 4—12 hours in a cool place before acquisition and inoculation feedings. Young aphids were used in groups of 5 or 10 per test plant according to the size of the plant.

Electron microscope was mainly used to confirm an infection with unclear symptoms. Virus preparations were made by dip method or after glutaraldehyde method of Lesemann (1972). The last-mentioned method was useful especially for carnation viruses.

The thermal inactivation point was determined by heating 2 ml lots of undiluted fresh sap from infected plants for 10 minutes at different temperatures.

Fresh sap from carnation, *C. quinoa*, *S. vaccaria*, *S. armeria* inoculated by carnation viruses was tested against various antisera in agar gel double diffusion tests (Ouchterlony test, cf. Kado and Agrawal 1972). In the case of chrysanthemum viruses, sap from *N. clevelandii*, *P. hybrida*, or chrysanthemums was used. As controls the antisera were tested against sap from healthy plants.

In tests with carnations, use was made of antisera to carnation mottle, vein mottle, latent and ringspot viruses and in a lesser extend, also of antiserum to necrotic fleck virus. For chrysanthemums use was made of antisera to the B-virus and the aspermy virus.

## RESULTS

## I Carnation viruses

Four viruses occurred in samples from imported cuttings and five in samples from farmers' stocks. The symptoms in test plants and some other properties of these viruses are described here. Special attention was

paid to the symptoms in S. vaccaria and S. armeria.

Carnation mottle virus, CaMV

The host range and symptoms of CaMV are reported by several authors (Kassanis 1955,

Brierley and Smith 1957, Hollings and Stone 1964, Åhman 1969). CaMV has been found earlier in Finland (Bremer 1978). In our tests it infected *C. amaranticolor*, *D. barbatus*, and *G. globosa* and showed symptoms similar to those reported by above mentioned authors.

C. quinoa was the most suitable test plant for CaMV. CaMV and CaVMV occurred nearly always together, which greatly affected the symptoms. When only CaMV infected C. quinoa, local spots appeared in 4—9 days, in summer more slowly than in winter. Yellow spots, 2—3 mm in diameter, turned red after a week. When spots were abundant, the whole leaf withered. Systemic symptoms, consisting of yellow spots, distortation and stunting of the leaves and dwarfing of the whole plant, appeared 12—14 days after inoculation.

S. vaccaria showed local vein chlorosis and mottle, sometimes yellow spots in 7—10 days. 10—14 days after the infection systemic chlorotic flecks, distortation and curling of the leaves and dwarfing of the plants began to appear.

S. armeria showed systemic mottling and curling of leaves.

## Serological test

In agar gel double diffusion tests the mottle virus in crude sap of *C. quinoa* or *D. barbatus* reacted positively with homologous antiserum.

Carnation vein mottle virus, CaVMV

## Symptoms in test plants

CaVMV induced in *C. amaranticolor* small chlorotic and necrotic local spots, about 1 mm in diameter, which might later have red centers. No systemic symptoms appeared. In-

fected *C. quinoa* plants were sometimes symptomless. Sometimes they showed chlorotic local spots in 7—10 days. Spots tended to appear along the veins. Systemic symptoms, consisting of distortion and curling of the leaves, appeared in 2—4 weeks (Fig. 4). The above symptoms were very strong when the *C. quinoa* plants were infected with both CaMV and CaVMV.

S. vaccaria showed local symptoms as when infected by CaMV, but much stronger. About two weeks after inoculation the plants developed systemic vein chlorosis, deformation of the leaves and dwarfing of the plants. The flower clusters curled into tight bundles and the flower stalks bent strongly downwards. Corollas were dwarfed and deformed. The number of the petals was more than in normal flowers. The petals were also grouped asymmetrically in flowers. Colour breaking and narrowing of the petals occurred often, too (Fig. 5).

The symptoms described above are similar to those described by other workers (Kassanis 1955, Hakkaart 1964, Hollings et al. 1977).

#### Serological test

The identification of CaVMV was confirmed in agar gel double diffusion tests using sap from infected *C. quinoa* and *S. vaccaria*. CaVMV reacted strongly positively with the antiserum.

# Electron microscopy

In the dip preparations made from leaves of S. vaccaria and C. quinoa a few long flexuous particles were seen under the electron microscope. Few particles could be measured, their length varying from 680—740 nm, but reliable measurements could not be obtained because of the small number of particles.

Carnation etched ring virus, CERV

Symptoms in test plants

To detect CERV we mainly used *S. armeria* and *S. vaccaria*, the last one being very suitable.

#### S. vaccaria

Plants infected at an early stage developed a very marked disease and usually died. The symptoms varied greatly according to the season. A very typical symptom was development of local and systemic, light brown, necrotic, irregular lesions with sharp borders. Only a few lesions per plant appeared. Necrotic lesions appeared in the leaf blade in 9-33 days. Lesions tended to locate near the main vein in the basal part of the leaf. From here the leaf began to wither. Later the whole leaf, sometimes even the whole plant, withered, especially in mixed infections. Besides necrotic lesions, grey or reddish concentric rings or yellow spots might develop in the inoculated leaves. At least a month after the inoculation, shortening of the internodes followed by dwarfing of the plants was observed.

#### S. armeria

After sap inoculation only a few plants became infected, and they showed faint symptoms. If the plants were infected with both mottle and etched ring viruses, the symptoms were clear and strong.

S. armeria was suitable as a test plant only in its vegetative phase, in winter time. Then CERV induced systemic symptoms consisting of bright white stripes and rings (Fig. 1). The breaking stripes often appeared on the midrib and veins, building a netlike figure. La-

ter the dwarfed plants abundantly formed sideshoots and branches and became bushy.

Thermal inactivation point

When the sap of infected S. armeria plants was heated at 80°C (for 10 minutes) a little infectivity was retained, but when heated at 85°C the infectivity was nil. S. armeria and S. vaccaria were used as test plants in these tests.

## Aphid transmission

It was possible to transmit CERV from infected S. armeria plants to healthy S. armeria plants in a non-persistent manner.

With regard to symptoms, thermal inactivation point and transmissibility through *M. persicae* aphid, CERV described above is similar to those isolated by other workers (cf. Hollings and Stone 1961, Kristensen 1963, Hakkaart 1968, Åhman 1969, Paludan 1970).

Carnation ringspot virus, CRSV

Symptoms in test plants

#### G. globosa

Small, light necrotic rings and lesions appeared in the inoculated leaves 3—4 days after the inoculation. Large irregular systemic lesions followed by mottling, reddening and deformation of the leaves appeared in a week.

#### N. clevelandii

Dark grey, necrotic local spots 1,5 mm in diameter appeared in 3 days in the inoculated leaves, which died within 10 days after the inoculation. Systemic symptoms were very strong. New leaves were dwarfed, chlorotic



Fig. 1. Systemic symptoms of carnation viruses on the leaves of *S. armeria*: on the right the symptoms of etched ring virus and in the middle the symptoms of mottle virus. The leaf on the left is healthy.



Fig. 2. Local symptoms of carnation ringspot virus on the leaves of *S. vaccaria*. Two leaves on the right are healthy.



Fig. 3. On the right a carnation leaf, from which the necrotic fleck virus was isolated.



Fig. 4. Systemic yellowish mottling, buckling and distortion caused by carnation vein mottle virus in *C. quinoa*. Unioculated control on the left.



Fig. 5. On the right a flower of *S. vaccaria* infected with both carnation vein mottle and mottle viruses showing dwarfed and narrow petals.

and crinkled. The whole plant dwarfed and many infected plants died inside three weeks.

## S. vaccaria cv. Pink Beauty

Greyish ring spot appeared in inoculated leaves in three days (Fig. 2). Systemic symptoms appeared a week after the inoculation, consisting of mottling, vein chlorosis, necrosis and deformation of the leaves and dwarfing of the plants.

#### S. armeria

Local greyish white rings and stripes along the veins appeared 3—4 days after the inoculation. Systemic symptoms, white small spots and streaks appeared after a week.

## Thermal inactivation point

When the sap from infected N. clevelandii plants was heated to  $+82^{\circ}\mathrm{C}$  only a few N. clevelandii or C. amaranticolor plants became infected. When heated to  $+85^{\circ}\mathrm{C}$  no plants became infected.

#### Serological test

In an agar gel double diffusion test CRSV in crude sap of *C. quinoa* reacted positively with the antiserum for ringspot virus.

The CRSV described here seems to be similar to that described in the literature (cf. Kassanis 1955, Brierley and Smith 1957, Hakkaart 1964, Hollings and Stone 1965, 1970).

## Carnation necrotic fleck virus, CNFV

CNFV, which is difficult to transmit with sap and which infects only *Dianthus* and relative species (Inouye and Mitsuhata 1973) could not be detected in the routine tests,

because of those properties. But in electron microscope examinations long flexuous particles like those of CNFV (cf. Inouye and Mitsuhata 1973) were found abundantly (Fig. 6). Particles were found in glutaraldehyde preparations made from samples taken from farmers' cultivations, from cvs. Calypso, Pallas and an unknown cv. The length of the particles varied from 650 to 1500 nm, being on average 1183 nm. In total 298 particles were measured, 265 of which had a length of 1150—1350 nm (Fig. 7), the width being 13—15 nm.

Those carnation plants inspected under electron miscroscope, showed many different symptoms, such as chlorotic flecks, mottling and streaking on the leaves (Fig. 3). According to tests with test plants and with different antisera these carnation plants were infected with CaVM, CaVMV and CERV, too. CaMV and CERV viruses have spherical particles (Kassanis 1955, Lawson et al. 1977) but CaVMV has long flexuous particles. According to Weintraub and Ragetli (1970) their length varied from 675 to 850 nm in purified sap and the longest particles in thin section were about 750 nm. The number of CaVMV particles was small.

The abundance of the longer particles in the present examinations suggests that CNFV was also infecting carnations. Workers have got somewhat different results from the measurement of CNFV particles. According to Inouye and Mitsuhata (1973) particles were  $1\,400-1\,500\,$  nm  $\times\,12-13\,$  nm, according to Smookler and Loebenstein (1974) about  $1\,250\,$  nm  $\times\,13\,$  nm, and according to Mayhew (1979)  $1\,160\,$  nm  $\times\,12\,$  nm. Our measurements agree with those made by Mayhew (1979) and Smookler and Loebenstein (1974).

Aphid and sap transmissions to isolate the virus were done from carnation plants in which virus particles were detected.

To avoid infecting the test plants with the non-persistent CaVMV virus which often oc-

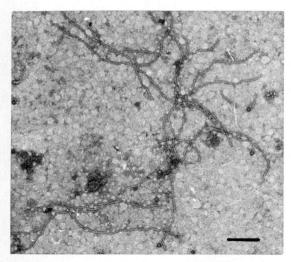


Fig. 6. Particles of carnation necrotic fleck virus. Preparate is made with glutaraldehyde, stained with PTA. Scale bar = 300 nm.

curred in the same plants, and which is also transmitted by M. persicae, the following method was used. Half an hour after acquisition feeding, the same aphids were transferred every two hours to new test plants (S. armeria and D. barbatus). A further transmission from those S. armeria plants showing symptoms was made by aphids to D. barbatus, D. chinensis, D. deltoides, D. plumarius, S. armeria and S. vaccaria. These plants showed symptoms which are described below. Sap transmission from plants infected with aphids was done to the test plants of same species, but did not succeed well. Most plants did not show any symptoms. The presence of the virus in plants inoculated by aphids or by sap was confirmed by electron microscope inspections.

#### Symptoms in test plants

## D. barbatus

In 2—3 weeks after the aphid transmission, vein chlorosis and necrosis appeared in young

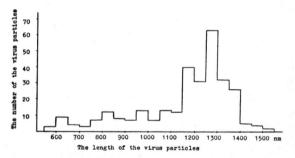


Fig. 7. A diagram of the length of the CNFV particles found in electron microscope preparations made from three various cultivars of carnation.

leaves, forming netlike figures. Leaves often reddened and their tips withered. Later the symptoms developed only in the parts of the leaf near the tip. When plants of *D. barbatus* were inoculated with sap, local pale necrotic spots appeared in a few of the inoculated plants in 10 days.

#### S. vaccaria

Faint lesions appeared in a week and during the next week developed into necrotic lesions. Systemic symptoms consisting of mottling of the youngest leaves and flowers was observed in plants which were infected also with vein mottle virus.

## S. armeria

White flecks and streaks, very similar to those caused by etched ring virus, appeared on the leaves in 6—8 weeks after the aphid inoculation. The flecks tended to follow the veins. Sometimes dark purple flecks or reddening of the leaf boarders occurred.

## D. deltoides

3—4 weeks after the inoculation through *M. persicae* aphids, white spots and crinkling

of petals appeared in the flowers. The leaves were very narrow and curled, and whole plants dwarfed.

#### D chinensis

Three weeks after the inoculation through aphids, systemic pale spots appeared on the leaves. Later the centres of the spots turned reddish, and yellow mottling and flecking appeared on the crinkled leaves. White spots and flecks appeared in petals in flowers.

#### D. plumarius

The leaves had pale flecks, and plants were strongly dwarfed. White flecks appeared in petals.

## Serological test

To confirm the virus identification some serological tests were done. An antiserum for CNFV was used in double diffusion test against sap from carnations and *S. armeria* and *S. vaccaria*. The results were not reliably clear, though a faint precipitation line appeared in a few cases.

#### II Virus diseases of chrysanthemum

Samples of chrysanthemum cuttings were much healthier than those of carnation. Only two virus diseases and a viroid disease occurred in chrysanthemum cuttings and in samples taken from farmers' stocks.

#### Chrysanthemum virus B, CVB

This virus was isolated from chrysanthemums by sap inoculation to *Petunia hybrida*. Three cultivars, viz. Cream Star, Himmelröschen and Resisto Rosa, were used as test plant. Later only »Resisto Rosa» was used, because only it showed the symptoms of CVB clearly.

Round, yellow spots with distinct boarder lines appeared in 2—4 weeks in the inoculated leaves. At first the spots were small, 1—2 mm in diameter, but they grew, being later 0,5 cm in diameter. These spots appeared only in the inoculated, fully developed leaves. No systemic symptoms were observed in *P. hybrida* in the present tests, though according to Hollings (1957) CVB might be latent in the young leaves. The season was of importance to the development of the symptoms. In March—June the local spots appeared in *P. hybrida* in 10—15 days but in October—January only 24—29 days after the inoculation.

*N. clevelandii* was a reliable indicator for CVB. Systemic symptoms, consisting of vein clearing and mottling of the leaves appeared 2,5—3 weeks after the inoculation.

#### N. glutinosa

Few chlorotic local lesions appeared in 2—3 weeks. Systemic symptoms developed very seldom.

# Thermal inactivation point

When the sap from infected *N. clevelandii* or *P. hybrida* plants was heated for ten minutes some infectivity was retained at 70°C but none at higher temperatures. *N. clevelandii* and *P. hybrida* were used as test plants, too. This thermal inactivation point agrees with that determined by Hollings and Stone (1972). They report it to be 70—75°C for the sap from chrysanthemums and for 75—80°C for the sap from *P. hybrida*.

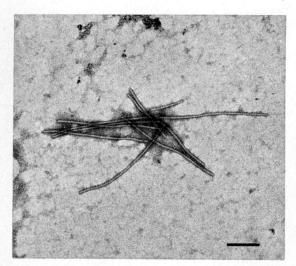


Fig. 8. Particles of chrysanthemum virus B. A dip preparate, stained with PTA. Scale bar = 200 nm.

## Serological test

An antiserum against CVB was used in agar gel double diffusion tests against sap from infected *P. hybrida*, but usually no precipitation lines were formed, even when the presence of the virus in *P. hybrida* was detected by the reaction of test plant and electron microscope examination.

#### Electron microscopy

Examination of dip preparations revealed similar, rather straight rods in all preparations from sap of infected  $P.\ hybrida,\ N.\ clevelandii$  and chrysanthemum, cv. Mistletoe (Fig. 8). The mean length of 100 particles from  $N.\ clevelandii$  was 706 nm and the width 12—13 nm. This agrees well with the results of other workers. After the first measurement made by Noordam (1952) the size of particles was  $600\times30$  nm, but according to Hollings and Stone (1967) the length of the particles was 700 nm in dip preparations. After Stone et al. (1970) the size of the particles was  $685\times12$  nm in purified extracts.

Chrysanthemum stunt viroid, CSV

The only used test plant for CSV was *C. morifolium*, cv. Mistletoe. First systemic symptoms, pale yellow spots and crinkling of the leaves appeared about three weeks after grafting. In 7—9 weeks faint vein chlorosis, yellowing of the leaves and yellow flecks of irregular shape and about 3—6 mm in diameter appeared. Later leaves and whole plants stunted.

# Chrysanthemum aspermy virus, CAV

Two CAV isolates were obtained from chrysanthemum cuttings. They caused principally similar symptoms, but the one caused severe symptoms and the other one mild ones in test plants. The isolates were detected by their symptoms in *P. hybrida*.

P. hybrida cv. Resisto Rosa. CAV induced local symptoms, round yellow flecks and yellow vein banding about 10 days after the inoculation. After 15—20 days systemic symptoms occurred consisting of dark green and yellow green mosaic, colour breaking and deformation of the flowers, crinkling and narrowing of the leaves. The narrowing of the leaves was often so intense that about a third of the tip part of the leaf consisted merely of the midrib.

Besides P. hybrida our CAV isolates caused typical CAV symptoms in C. amaranticolor, N. tabacum Samsun and White Burley, N. glutinosa, N. clevelandii, Cucumis sativus L. and Lycopersicum esculentum Mill. These symptoms were similar to those described by other authors (cf. Blencowe and Caldwell 1946, 1949, Hollings and Stone 1971).

The thermal inactivation point was determined by heating sap of infected *N. tabacum* Samsun using *N. glutinosa* and *N. clevelandii* as test plants. Sap of the isolate with strong symptoms infected a few plants when heated

at  $65^{\circ}$ C but none at  $70^{\circ}$ C. Sap of the isolate with mild symptoms had some infectivity when heated at  $60^{\circ}$ C but lost it at  $65^{\circ}$ C.

Occurrence of the virus diseases in carnation and chrysanthemum cuttings

Results from virus tests on carnation cuttings are summarized in Table 1. About half the cuttings were infected on arrival in Finland though the cuttings did not show any symptoms. CaMV and CaVMV, which usually occurred together, were the most common viruses in cuttings. In 1978, when no serological testing with CaVMV was done, there was no reliable record on the occurrence of CaVMV virus. CaMV symptoms in C. quinoa often covered the symptoms of CaVMV. CERV occurred abundantly, too.

There was a difference in health among cuttings from different origin. Cuttings propagated in Teneriffa or in the Federal Republic of Germany were very healthy, whereas Swedish cuttings propagated in Portugal were heavily virus infected. So were the French cuttings. Diseased cuttings had mixed infections of CaMV, CaVMV and CERV. Cuttings from Holland were free of CaMV and CaVMV but were very often infected with CERV.

In addition to the samples from imported cuttings, 27 carnation samples collected from

Table 1. Virus diseases in carnation samples of imported cuttings on average 1978—1979.

the number	infected	CaMV	CaVMV	CERV	CRSV
of the samples	%	%	%	%	%
93	51,4	39,7	40,81	29,1	1,1

<sup>1</sup> only from the year 1979

farmers' cultivations were virus tested. They were all infected with CaMV and nearly all with CaVMV and CERV. In the carnation samples from farmers, a virus which was very probably CNFV was found too. It was not possible to find out how common it was in imported cuttings, because our test methods were not suitable for detection of CNFV. But the abundant occurrence of virus particles in several carnation samples from three cvs. suggests that CNFV might be quite common in cuttings.

 $18,2\,^{9/0}$  of the imported chrysanthemum cuttings were virus infected. They were infected mainly by chrysanthemum virus B and a couple of samples by aspermy virus and stunt viroid.

Besides samples from cuttings, 10 samples were also taken from farmers' stocks. Besides CVB, CSV and CAV each occurred in one sample.

#### DISCUSSION

CaMV seems to be the most common virus in carnations. Its infection is difficult to avoid because of the high infectivity and stability of this virus.

CaVMV occurred very abundantly, too. Hollings et al. wrote in 1977, that »the carnation vein mottle virus was then not a danger to carnation crops in Britain, but sending

carnation plants to overwinter outdoors in warmer countries involves risks of more rapid spread by *M. persicae.*» This seems to have happened. The abundant occurrence of CaVMV in cuttings produced in South-Europe, especially in Portugal, is very probably due to infection with aphids in outdoor propagation fields. *Myzus persicae* is an

efficient vector (Hollings et al. 1977) and it occurs on carnations in Portugal (Ilharco 1973). It is not known, how effective other aphids which occur on carnations in South-Europe, are as vectors of viruses. It is not difficult to free carnation plants from CaVMV with heat therapy and meristem culture (Brierley 1964, Stone 1968, Hollings et al. 1977) but it seems to be difficult to prevent the reinfection through aphids.

CERV is not easy to detect serologically because of its low concentration in plants, and the test plants used earlier have not given reliable results (cf. Hollings and Stone 1969). Nowadays S. vaccaria seems to be a rather good indicator. CERV is difficult to eliminate from carnations with heat therapy (cf. Brierley 1964, Hollings 1967, Hakkaart and Jordanova 1968, Paludan 1970). CERV is transmitted by M. persicae, too. These two properties are probably the reason why this virus was quite common in the cuttings.

Ringspot virus was detected in one sample only. The virus is easily eliminated with heat therapy and meristem culture (cf. Kassanis 1955, Hollings and Stone 1965, 1970), and it has been eradicated from Dutch stocks (Ten Hoyten et al. 1968).

Carnation latent virus, which should be detectable in *C. quinoa* or by serological test or electron microscope, was not found in the cuttings nor in the samples taken from farmers' stocks, allthough all the above test methods were used.

A virus similar to necrotic fleck virus occurred abundantly in all inspected plants of three cultivars in a farmers cultivations. There were no aphids in these glasshouses. These three cultivars originated in cuttings of mother plants grown in Portugal, so this virus too has very probably come with cuttings.

The imported chrysanthemum cuttings were quite healthy. Only occasional infections of CVB, CAV and CSV occurred. Besides CSV, another viroid, chrysanthemum chlorotic mottle, infects chrysanthemums. This viroid, which causes symptoms in »Deep Ridge» chrysanthemums (Romaine and Horst 1975), was not found in the present tests. Most of the chrysanthemum cuttings in 1978---1979 originated from certificated material. This might explain why the chrysanthemum cuttings were virus-free compared with the carnation cuttings.

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## SELOSTUS

#### Virustaudit maahantuoduissa neilikan ja krysanteemin pistokkaissa

KATRI BREMER ja MARJA-LEENA LAHDENPERÄ

## Maatalouden tutkimuskeskus

Viljelijöiden ja pistokkaiden maahantuojien välillä on ollut erimielisyyttä neilikka- ja krysanteemiviljelyksillä esiintyneiden virustautien alkuperästä. Tämän vuoksi testattiin vuosien 1978—1979 aikana yhteistoiminnassa Kasvintarkastustoimiston kanssa maahan tuotujen neilikan ja krysanteemin pistokkaiden virustautisuutta. Lisäksi tarkastettiin viljelijöiden kasvihuoneista otetut 27 neilikka- ja 10 krysanteeminäytettä. Testaukset suoritettiin pääasiallisesti ilmaisinkasvien avulla,

mutta seerumitestauksia ja elektronimikroskopointia käytettiin myös apuna.

Maahantuontieristä otetuista neilikanpistokkaista oli 51,4 % viroottisia, viljelijöiltä otetut näytteet olivat kaikki viroottisia. Krysanteemin pistokkaat olivat sangen terveitä, 203 näytteestä 18,2 % oli viroottisia. Viljelijöiltä otetuista 10 näytteestä 7 oli viroottisia.

Neilikan virukset esiintyivät useimmiten sekainfektioina. Yleisimmät virukset olivat läikkävirus ja suoniläikkävirus, jotka v. 1979 infektoivat yhdessä 40,8 % näytteistä. Läikkäviruksen infektoimia näytteitä oli edellisenä vuonna 38,6 % kaikista näytteistä.

Neilikan valkokirjovirus osoittautui yllättävän yleiseksi v. 1978 29,5 % ja v. 1979 28,6 % näyteeristä todettiin tämän viruksen saastuttamiksi. Valkokirjoa tavattiin nimenomaan sellaisissa näytteissä, joissa ei esiintynyt läikkä- eikä suoniläikkävirusta. Neilikan rengaslaikkuvirusta tavattiin vain yhdestä näytteestä. Neilikan nekroosilaikkuviruksen kaltaisia virushiukkasia löydettiin elektronimikroskoopin avulla muutamista viljelijöiltä otetuista näytteistä. Tämä virus ei siirry helposti mehussa, eikä sen vuoksi voinut tulla esilleilmaisinkasvitesteissä. On varsin todennäköistä, että tämäkin virus on tullut pistokkaiden mukana. Pelkästään kirvojen avulla leviten se ei olisi ehtinyt tulla viljelyksellä niin yleiseksi kuin se Neilikan nekroosilaikkuviruksen on vasta äskettäin todettu esiintyvän Euroopassa (Rana ym. 1977) ja sen vuoksi sen torjuntaankaan ei ole kiinnitetty vielä huomiota.

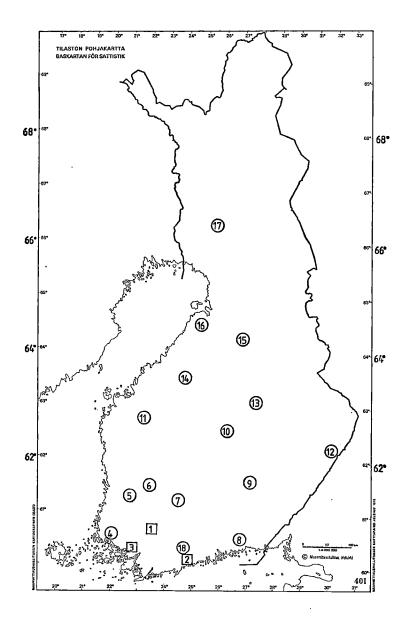
Neilikan pistokkaiden alkuperään nähden oli eroja viroottisuudessa. Teneriffalla ja Länsi-Saksassa tuotetut pistokkaat osoittautuivat erittäin terveiksi, kun sen sijaan ruotsalaiset pistokkaat, jotka oli lisätty Portugalissa, olivat hyvin viroottisia. Myös ranskalaiset pistokkaat olivat virustautisia. Monissa erissä ilmeni läikkä-, suoniläikkä- ja valkokirjoviruksen sekainfektio. Läikkävirus leviää helposti suuren tarttuvuutensa ja ulkoisten olosuhteiden kestokykynsä takia. Suoniläikkä- ja valkokirjovirus leviävät helposti kirvojen avulla emokasveihin pistokkaita avomaal-Suoniläikkävirus ei ole ollut la tuotettaessa. yleinen aikaisemmin, jolloin pistokkaita lisättiin kasvihuoneissa (vrt. Hollings ym. 1977). Hollannista tuoduissa pistokkaissa esiintyi valkokirjoa. Valkokirjovirus on vaikea poistaa neilikoista lämpökäsittelylläkään, joten se pysynee helposti neilikan emokasveissa.

Krysanteemin pistokkaissa esiintyy pääasiallisesti B-virusta, jonka aiheuttama tauti on yleensä lievä. Parissa näytteessä todettiin martovirusta ja viroidin aiheuttamaa kääpiökasvutautia. Aivan samat taudit esiintyivät myös viljelijöiltä saaduissa näytteissä. Eräällä viljelyksellä kääpiökasvutauti esiintyi sangen runsaana.

Krysanteemin pistokkaiden vähäinen virustautisuus johtunee siitä, että vuosina 1978—79 pistokkaita tuotettiin pääasiallisesti maista, joissa tervetaimituotannon ansiosta käytettiin puhtaita emokasveja.

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