Annales Agriculturae Fenniae

Research note

THE BLACKCURRANT VARIETY 'MORTTI'

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HIETARANTA, T. and HIIRSALMI, H. 1990. The blackcurrant variety 'Mortti'. Ann. Agric. Fenn. 29: 159—163. (Agric. Res. Centre, Inst. Hort., SF-21500 Piikkiö, Finland.)

The blackcurrant variety 'Mortti', originating from the crossing 'Öjebyn' x 'Wellington XXX', was released for cultivation in 1989. 'Mortti' grows a vigorous, high and rather erect bush. Its yield and berry size have in trials been equal to 'Öjebyn'. The variety has wintered very well in Southern Finland and its resistance to American gooseberry mildew has been satisfactory.

Index words: blackcurrant variety, *Ribes nigrum,* small fruit breeding.

INTRODUCTION

After mechanical harvesting became general in commercial blackcurrant cultivation many of the old cultivars, such as 'Brödtorp', 'Lepaan musta' (= 'Lepaa black') etc., were abandoned because of their spreading growth habit. At present, the selection of cultivars in Finland is very limited and the Swedish 'Öjebyn' is almost the sole cultivar used by professional growers. In Finnish blackcurrant breeding in addition to winterhardiness, resistance to *Spaerotheca mors-uvae,* high productivity and good fruit quality, improvement of the growth habit has been one of the main aims (HIIRSALMI 1988).

CROSSING AND EVALUATION

A crossing between the Swedish local variety 'Öjebyn' and the English 'Wellington XXX' was made in 1967 and it produced a very promising selection 67008081. On the basis of preliminary evaluations, this selection was picked out on the grounds of its heavy crop and desirable growth habit.

The cultivation value of selection 67008081 was tested in two trials. In the preliminary ob-servation trial, set up in 1975, 67008081 was compared both with the control variety 'Bröd-torp', and also with 'Öjebyn', a row of which grew just beside the trial. The selection seemed to be very promising, since its crop was heavi-er than either 'Brödtorp' or 'Öjebyn'.

A comparative trial was set up in 1982. Four bushes of each selection and the control varie-ty 'Öjebyn' were planted on loamy clay soil

using planting distances of 1 m in the row and 3 m between the rows. The observations and evaluations represented started in 1984. The beginning of flowering, and the time of leaf abscission were recorded. Vegetative characters, i.e. winter hardiness, mildew resistance, vigour and growth habit were visually evaluated using a scale of 0—10. Plants completely winter hardy, resistant, very vigorous and erect were graded 10. Fruit characters were evaluated using scales of 1—3 for firmness and the thickness of fruit skin, 1—5 for the blackcurrant aroma and 1—4 for sourness. Thus higher values indicate firmer fruit, thicker skin, stronger aroma and more sour taste. All evaluation results are given as means calculated from the yearly results for 1984—1987; the yield results are from the years 1985—1987. These are stated as the annual means of the bushes and standard errors of the means calculated from these.

RESULTS

Compared to 'Öjebyn', the yield and fruit size of selection 67008081 are about equal (Table 1), but the bushes of 67008081 are more vigorous and more erect in their growth habit. The

Table 1. Blackcurrant comparative trial planted in 1982, results in 1985—1987.

Variety	Yield kg/bush/year, $\bar{x} \pm$S.E.			Weight of 100 berries g, $\bar{x} \pm$S.E.		
	1985	1986	1987	1985	1986	1987
67008081	2.39±0.25	1.39±0.39	1.00±0.29	70.0±2.9	67.0±2.9	80.8±2.2
'Öjebyn'	2.16±0.18	1.44±0.03	1.16±0.11	92.8±3.8	58.5±2.1	67.0±0.8

Table 2. Vegetative and flowering characteristics of blackcurrants in comparative trial in 1984—1987.

Variety	Beginning of flowering Days from May 1 $\bar{x} \pm$S.E	Time of leaf abscission Days from October 1 $\bar{x} \pm$S.E.	Winter hardiness 0—10	Mildew resistance 0—10	Growth habit 0—10	Vigour 0—10
67008081	24.0±4.0	34.0±7.0	9.4	9.7	7.6	8.5
'Öjebyn'	22.0±3.0	26.7±4.9	9.1	10.0	6.2	7.7

Winter hardiness: 1—10 = dead — no winter injury
Mildew resistance: 0—10 = heavily infected — completely resistant
Growth habit: 0—10 = very spreading — very erect
Vigour: 0—10 = dead — very vigorous

Table 3. Fruit quality of blackcurrants in comparative trial in 1984—1987.

Variety	Firmness 1—3	Thickness of skin 1—3	Aroma 1—5	Sourness 1—4
67008081	2.6	2.1	3.4	2.7
'Öjebyn'	1.6	1.7	3.1	2.3

Firmness: 1—3 = tender — firm
Toughness: of skin 1—3 = thin — thick
Aroma: 1—5 = mild — strong
Sourness: 1—4 = weak — very sour

beginning of flowering occurs about 2 days and the leaf abcission about a week later than in 'Öjebyn'. The selection has wintered very well in Southern Finland, despite the occurrence of two exceptionally hard winters during the comparative trial, in 1984—1985 and in 1986—1987. The resistance of 67008081 to American gooseberry mildew has been satisfactory in field conditions (Table 2).

The fruits of 67008081 are fairly firm and their skin is thick, compared to the fruits of 'Öjebyn'. The aroma is also slightly stronger and taste slightly more sour than that of 'Öjebyn' (Table 3).

THE BLACKCURRANT VARIETY 'MORTTI'

The new blackcurrant variety 'Mortti' was released for cultivation in 1989. It originates from a crossing between the Swedish local variety 'Öjebyn' and the British 'Wellington XXX', which was made at the Institute of Horticulture in 1967. 'Mortti' grows a vigorous and high bush, usually about 130—150 cm. The growth habit of 'Mortti' is rather erect and in this respect it resembles 'Wellington XXX' more than 'Öjebyn'. The leaves of 'Mortti' are very much like the leaves of 'Öjebyn' i.e. the overall form is about the same and the leaf sinus is deep. The flowering takes place about two days and the leaf abscission about a week later than in 'Öjebyn'. The flowers are medium sized or big, and light with some reddish stripes. 'Mortti' has been winterhardy in Southern Finland and its resistance to American gooseberry mildew has been satisfactory.

The yield and fruit size of 'Mortti' have been equal to 'Öjebyn'. The average weight of 100 berries has been between 70 and 105 grams. The crop ripens a few days later than the crop of 'Öjebyn'. The blackcurrant aroma of the fruits is medium strong. The fruits are rather firm and their skin is relatively thick. The comments received from industry indicate that the berries of 'Mortti' are suitable for industrial use; their taste is good and their aroma is satisfactory. They can be used for syrups and jams but also for fresh use.

Because of its erect growth habit and firm fruit, 'Mortti' seems to be suitable for mechanical harvesting, and it is recommended for trials for professional cultivation.

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SELOSTUS

Mustaherukkalajike 'Mortti'

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Maatalouden tutkimuskeskus

Mustaherukkajalostetta 67008081 risteytyksestä 'Öjebyn' ja 'Wellington XXX' verrattiin 'Brödtorp' ja 'Öjebyn'-lajikkeisiin. Jaloste todettiin viljelyarvoltaan 'Öjebyn'-lajikkeen veroiseksi ja se laskettiin viljelyyn vuonna 1989 lajikenimellä 'Mortti'. 'Mortti' muodostaa korkean, voimakaskasvuisen ja pystyn pensaan. Pystykasvuisuuden suhteen se muistuttaakin enemmän 'Wellington XXX' kuin 'Öjebyn'-lajiketta. Lehdiltään 'Mortti' on puolestaan suuresti 'Öjebyn'-lajikkeen kaltainen. 'Mortti' kukkii paria päivää ja tuleentuu noin viikkoa myöhemmin kuin 'Öjebyn'. Keskikokoiset tai suuret kukat ovat vaaleita, mutta niissä on punertavia juovia. Lajike on talvehtinut Etelä-Suomessa erittäin hyvin ja se on kenttäolosuhteissa osoittautunut riittävän härmänkestäväksi.

Satoisuudeltaan ja marjakooltaan 'Mortti' on kokeissa ollut 'Öjebyn'-lajikkeen veroinen. Keskimääräinen sadan marjan paino on kokeissa ollut kasvupaikasta ja vuodesta riippuen 70 ja 105 gramman välillä. Sato kypsyy hieman 'Öjebyn'-lajikkeen satoa myöhemmin. Marjojen aromi on mustaherukalle luonteenomainen ja keskinkertaisen voimakas. Marjat ovat mustia, kiinteitä ja niiden kuori on paksuhko. Ne ovat kohtalaisen tiukasti kiinni tertuissa. Teollisuus on arvioinut marjojen soveltuvan teolliseen käyttöön; niiden maku on hyvä ja tuoksu tyydyttävä. Marjoista voidaan valmistaa mehuja ja hilloja, mutta niitä voidaan käyttää myös tuoreena.

Pystykasvuisuutensa ja kiinteiden marjojensa perusteella 'Mortti'-mustaherukkalajike soveltunee konekorjuuseen, ja sitä suositellaan kokeiltavaksi ammattiviljelyyn.

Figs. 1 and 2. The variety 'Mortti'

Research note

THE STRAWBERRY VARIETY 'MARI'

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HIIRSALMI, H. and LAURINEN, E. 1990. The strawberry variety 'Mari'. Ann. Agric. Fenn.
29: 165—168. (Agric. Res. Centre, Inst. Hort., SF-21500 Piikkiö, Finland.)

In 1988, a strawberry variety produced by crossing 'Pocahontas' with 'Lihama' was
released for cultivation under the name 'Mari'. In trials it has produced a heavy crop,
averaging around 130 kg/100 m^2 of commercially viable fruit, and has also demon-
strated other good qualities. Its fruit are large, firm, and slightly sour. The bushes
are tall and airy. Its resistance to grey mould and mildew is satisfactory or better.
Under Finnish conditions, 'Mari' is an early variety.

Index words: strawberry cultivars, strawberry breeding.

INTRODUCTION

Out of over a hundred different strawberry cul-
tivars tested in trials at the Institute of Horticul-
ture in Piikkiö, very few have been found reli-
able enough for cultivation under Finnish con-
ditions. Neither Central European nor Ameri-
can cultivars provide adequate results under
Northern temperatures and light conditions to
be commercially viable. An outstanding excep-
tion is the cultivar 'Senga Sengana', originally
bred in Germany, which has now been the ma-
jor commercial cultivar in Finland for over
three decades. The other major cultivars in Fin-
land are at present the Danish 'Zefyr', and the
Norwegian 'Jonsok'.

In order to develop strawberry cultivars spe-
cifically suited for Finnish conditions, a breed-
ing project has been under way at the Institute
of Horticulture since 1961, using both cross-
fertilization and self-pollination (HIIRSALMI
1969), with the aim of producing varieties reli-
able in cultivation, with fruit suitable for fresh
consumption, for deep-freezing, and for indus-
trial use. In 1984, a high-yield late-crop culti-
var developed on this project was released un-
der the name of 'Hiku', with fruit primarily
suitable for fresh consumption (HIIRSALMI and
SÄKÖ 1985, HIIRSALMI 1988), but work has also
been continuously in progress on the develop-
ment of a variety for the various methods of
early-crop cultivation.

In strawberry breeding at the Institute of Horticulture, the major focuses of attention have been the size of the crop, the size of the fruit, winterhardiness, and disease and pest resistance. More recently, closer attention has also been paid to the improvement of internal quality, and another feature to be investigated in the future will be suitability for mechanical harvesting.

On the basis of the observations to date, the most promising early strawberry selection for further investigation is a cross between the American cultivar 'Pocahontas' and the German 'Lihama', 65029001. In trials, both in the open field and under cover, this selection has yielded a heavy crop with large fruit (Tables 1 and 2). In several other respects, too, this selection is at least as advantageous as 'Zefyr', the most widely cultivated early cultivar, and its resistance to mildew is better. One factor adversely affecting the quality of the fruit, and thus also reducing the commercial yield, is the ridged shape, especially in the large fruit ripening first in the crop.

In view of its encouraging characteristics, selection 65029001 was released for sale in 1988, under the name 'Mari'.

Table 1. Results of the strawberry variety trials, 1979—81. Planting density 33 × 100 cm.

Variety	Total yield kg/100 m²			Proportion of crop			Weight of fruit g	Proportion of crop during first two weeks
	1979	1980	1981	Fit for sale kg/100 m²	Small %	Moulded %		
Hiku	111	216	259	170	6	7	14.2	58
Mari (= 65029001)	76	137	146	110	6	2	12.0	79
Zefyr	61	117	176	107	7	2	10.0	89
Senga Sengana	93	111	178	101	8	12	10.1	61
Hella	48	87	81	62	9	4	8.5	81

Table 2. Results of early strawberry variety trials in unheated greenhouse, under fibercloth cover, and in open fields, 1985—87. Planting density 33 × 70 cm.

Variety	Total yield kg/100 m²			Proportion of crop		Weight of fruit g	Proportion of crop during first two weeks
	1985	1986	1987	Fit for sale kg/100 m²	Small %		
In open fields							
Mari (= 65029001)	171	229	131	150	14	13.0	92
Jonsok	216	197	161	147	23	10.3	98
Zefyr	168	195	109	132	14	13.2	97
Riva	116	147	60	91	15	10.5	99
Under fibrecloth							
Mari (= 65029001)	170	214	120	131	20	11.0	73
Zefyr	130	172	127	112	21	10.0	84
Riva	74	141	78	76	23	9.9	97
In greenhouse							
Mari (= 65029001)	150	207	116	125	21	11.0	82
Jonsok	211	138	175	109	38	7.7	93
Zefyr	114	108	82	76	24	8.8	95
Riva	77	107	67	65	23	8.0	85

DESCRIPTION OF THE VARIETY 'MARI'

The strawberry cultivar 'Mari' is a selection from the progeny of a cross carried out at the Institute of Horticulture, Piikkiö, Finland, in 1965 between the cultivars 'Pocahontas' and 'Lihama'. It has inherited features from both parent stocks.

The plants are airy, with straight, thick leaf stalks. The leaflets are pale green, somewhat greyish on the underside, and medium to large in size. The flowers are formed partly under cover of the leaves, and are large. As the fruit ripen, the racemes bend out to the sides.

This cultivar has been found to winter well, both in South-West and Central Finland at least. Its resistance to grey mould and mildew is satisfactory or better. The proportion of moulded fruit in the crop has varied in trials, depending on seasonal and local temperature and moisture conditions, between 0 and 10 per cent.

'Mari' can be classified as an early variety.

The first fruit ripen on average two days earlier than in 'Zefyr', although the crop continues for longer than with 'Zefyr'.

The commercially viable crop obtained from 'Mari' in trials has been fair to good, averaging around 130 kg/100 m². It also appears to be better adapted than 'Zefyr' to cultivation under fibrecloth cover or in unheated greenhouses.

The fruit of 'Mari' are conical in shape, relatively flat, often ridged, and large. The mean size of fruit cultivated in open-field conditions is around 13 g. The petals sometimes remained attached to the calyx even after the fruit have ripened. The fruit are very firm, and bright red in colour, with somewhat paler flesh; the flavour is pleasant, mild, and slightly sour. The fruit are definitely suitable both for fresh consumption and for deep-freezing.

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SELOSTUS

Mansikkalajike 'Mari'

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'Mari'-mansikkalajike on valittu vuonna 1965 Maatalouden tutkimuskeskuksen puutarhatuotannon tutkimuslaitoksella tehdyn risteytyksen 'Pocahontas' x 'Lihama' jälkeläistöstä. Se muodostaa korkean, ilmavan kasvuston. Kukinta tapahtuu osittain lehdistön suojassa. Marjojen kypsyessä tertut taipuvat kasvuston sivulle. Lajikkeen talven-, harmaahomeen- ja härmänkestävyys ovat vähintään tyydyttävät.

'Mari' voidaan luokitella aikaiseksi lajikkeeksi. Sadonkor-juuajaltaan se on verrattavissa yleisesti viljeltävään 'Zefyr'-varhaislajikkeeseen. 'Mari'-lajikkeen myyntikelpoinen sato on kokeissa ollut tyydyttävä tai hyvä, keskimäärin noin 130 kg/100 m². Marjat ovat litteän kartiomaisia, usein harjuisia ja varsin kookkaita. Keskimääräinen marjapaino on ollut avomaalla noin 13 g. Marjat ovat varsin kiinteitä ja pintaväriltään heleänpunaisia, malloltaan vähän vaaleampia. Ne ovat miellyttävän makuisia, mietoja ja lievästi happamia.

Research note

THE RED RASPBERRY VARIETY 'VILLE'

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HIIRSALMI, H. and LAURINEN, E. 1990. The red raspberry variety 'Ville'. Ann. Agric. Fenn. 29: 169—171. (Agric. Res. Centre, Inst. Hort., SF-21500 Piikkiö, Finland.)

In 1988, a raspberry variety produced by crossing the 'Ottawa' cultivar with a natural strain originating from Hautjärvi, Mäntsälä, in southern Finland, was released under the name 'Ville'. In trials, the yield has been satisfactory, averaging 57 kg/100 m². The fruit are small, similar to the fruit of the wild raspberry, and have a fine aroma. The variety produces luxuriant, tall canes. Winterhardiness is good, and resistance to blight satisfactory. 'Ville' is suited to domestic production.

Index words: raspberry cultivars, raspberry breeding.

INTRODUCTION

The area under small fruit cultivation in Finland in 1988 totalled around 4 300 hectares, mainly for strawberries and currants (ANON. 1988), with only around 100 ha given over to raspberries. The low level of interest in raspberry cultivation is largely due to the fact that the available cultivars are very poorly adapted to Finnish climatic conditions (SÄKÖ and HIIRSALMI 1980). The cultivar most widely used at present is 'Ottawa', from Canada; two others in significant use are the Canadian cultivar 'Muskoka' and the German 'Preussen'. These frequently suffer winter damage, however, sometimes so badly that no crop is obtained at all. The yield is also often severely reduced by the effect of virus diseases and cane blight (*Didymella aplanata*). Previously, no cultivars bred within Finland have been available.

The wild raspberry *Rubus idaeus* L., occurs naturally throughout most of Finland. There are wide differences between the natural strains of rate and habit of growth, and in the yield and size of the fruit (ROUSI 1965, HIIRSALMI 1971, 1976). In field trials, natural strains collected from various parts of Finland have been found to be significantly more winterhardy than cultivars; the natural strains are unsuitable for commercial production, however, since their fruit are small in size and the yield is low.

In 1973, a raspberry breeding project was launched at the Institute of Horticulture in Piikkiö of the Agricultural Research Centre of Finland, with the aim of utilizing the genetic stock of natural strains (HIIRSALMI 1988). By crossing the best cultivars with natural raspberry strains displaying desirable features, it has proved possible to transfer valuable characteristics, in particular winterhardiness.

The main focuses of attention in the raspberry breeding project have been improved yield, size of fruit, resistance to blight, and, in particular, winterhardiness. One of the essential conditions for successful wintering is that the canes must ripen in good time before the first frosts in autumn. Increasing value has also been attached to the internal quality of the fruit. Other factors which need to be taken into account in breeding are the development of plants suitable for mechanical harvesting, which is essential for extensive commercial cultivation; the attachment of the fruit; and the timing of the ripening of the crop.

On the basis of initial observations, a number of hybrids between cultivars and between cultivars and natural strains have been selected for further investigation. The results so far show that selection 73059009, obtained by crossing the cultivar 'Ottawa' with a natural strain from

Hautjärvi, Mäntsälä, in southern Finland, is distinctly more winterhardy than any other (Table 1). This selection produces a heavy crop of small fruit, with a fine aroma, so that the total yield is satisfactory. Another selection, 73177003, obtained by crossing 'Ottawa' with 'Glen Cova', has also attracted attention. In normal years, this has produced a considerably higher yield than the existing cultivars; during the harsh winters of 1984—85 and 1986—87, however, it suffered serious damage, which severely reduced the yield. The fruit of this selection remain pale even when ripe, and have a somewhat unusual flavour.

Selection 73059009 has been shown to enjoy features not found in any existing cultivar, and was therefore released for cultivation in 1988, under the name 'Ville'. Selection 73177003 is under ongoing examination.

Table 1. Results of the raspberry variety trials, 1983—88. Planting density 70 × 300 cm.

Variety	Total yield kg/100 m²	Proportion Fit for sale, %	Weight per 100 fruit, g	Winterhardiness (1—9)
73177003	78	87	231	6.8
Muskoka	75	88	219	8.1
Ottawa	64	90	243	8.0
Festival	58	85	252	5.6
Ville (= 73059009)	57	84	144	8.3
Boyne	56	87	217	7.0
Haida	54	86	265	5.9
Carnival	46	87	183	7.1
Comet	41	84	183	7.1
Glen Cova	40	90	328	4.4
Glen Isla	35	87	303	4.3

Winterhardiness: 1—9 = all shoots dead — completely healthy
73059009 = 'Ottawa' × natural strain Mäntsälä/Hautjärvi
73177003 = 'Ottawa' × 'Glen Cova'

DESCRIPTION OF THE VARIETY 'VILLE'

The red raspberry variety 'Ville' is a selection from the progeny of a cross made in 1973 at the Institute of Horticulture, Piikkiö, Finland, between the cultivar 'Ottawa' and a natural strain from Hautjärvi, Mäntsälä, in the interior of southern Finland. The growth habit of the variety is a cross between those of the parent stocks. Its fruit are very similar to those of wild raspberry strains.

'Ville' produces luxuriant, tall canes with few

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thorns, distinctly thicker and taller than those of the wild raspberry strains, though slenderer than those of 'Ottawa'. The variery has wintered extremely well in southern Finland, with the exception of some damage to buds during exceptionally severe winters. Resistance to cane blight has been adequate.

The yield from 'Ville' has in normal years been satisfactory, averaging around 57 kg/100 m², as compared with 64 kg/100 m² for 'Ottawa'. The fruit are small, the weight of 100 fruit averaging 144 g, compared with 243 g for 'Ottawa', and 100 g for wild raspberry strains. The fruit are firm and have a fine aroma. Their tastiness makes them excellent for the manufacture of jam and juice, as well as for consumption while fresh. 'Ville' is particularly recommended for domestic cultivation.

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SELOSTUS

Vadelmalajike 'Ville'

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'Ville'-vadelmalajike on valittu vuonna 1973 Maatalouden tutkimuskeskuksen puutarhatuotannon tutkimuslaitoksella 'Ottawa'-lajikkeen ja Mäntsälän Hautjärveltä olevan luonnonkannan välillä tehdyn risteytyksen jälkeläistöstä. Se muodostaa rehevän, korkean kasvuston. Lajikkeen talvenkestävyys on hyvä ja versotaudinkestävyys tyydyttävä.

'Ville'-lajikkeen sato on kokeissa normaaleina vuosina ollut tyydyttävä, keskimäärin 57 kg/100 m². Sen marjat ovat pieniä, luonnonvadelman marjoja muistuttavia. Sadan marjan paino on keskimäärin ollut 144 g. Marjat ovat hyvin koossa pysyviä ja hienoaromisia. Maukkaina ne soveltuvat erinomaisesti hillon ja mehun valmistukseen sekä tuorekäyttöön. 'Ville'-lajiketta suositellaan etenkin kotitarveviljelyyn.

THE EFFECT OF SOIL COOLING ON THE FLOWERING OF SUMMER-PLANTED FREESIAS

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JUHANOJA, S. 1990. The effect of soil cooling on the flowering of summer-planted freesias. Ann. Agric. Fenn. 29: 173—178. (Agric. Res. Centre, Inst. Hort., SF-21500 Piikkiö, Finland.)

The effect of soil cooling on the timing of flowering of freesias was studied. The corms were planted in the first half of June and the soil was cooled by circulating cold water at about 8—10 °C in pipes placed in the peat. The effect was intesified with styrox grains as isolating material on top of the peat. From the beds with two cooling cycles and styrox grain cover, the first flowers were harvested 12—13 weeks after planting, while the same variety flowered 18—20 weeks after planting in beds without cooling and cover. One cooling cycle/bed was almost as effective as two cycles, but neither cooling nor cover alone was adequate to advance the flowering. There were six varieties in the trial; cooling and cover advanced the fastest-developing by 4—7 weeks, and the slowest-developing by up to 10 weeks. The duration of cooling varied from 6 to 12 weeks and six weeks was sufficient in most cases but a longer duration was not harmful either. A warm uncooled period of two weeks before the start of cooling, however, delayed the beginning of flowering by two weeks.

Index words: freesia, flowering, peat, soil cooling, styrox.

INTRODUCTION

After planting the corm-raised first develops a new corm, and from this some leaves, before flower induction. At a suitable temperature the flower is initiated after 7—9 leaves, and at that time the formation of leaves gradually decreases and ends. At temperatures too high the formation of leaves continues for longer. Flower induction is dependent on the temperature around the corm: at the optimum 13—15 °C, flower buds are formed in 6—10 weeks, whereas temperatures over 18 °C retard the development or prevent it (MANSOUR 1968). A temperature as constant as possible at about 15—16 °C is the best during the 6—10 first weeks after planting, because then the development of

leaves, the growth of flower stems and the flower induction are all at an optimum or nearly optimum stage (DIJKHUIZEN and van HOLSTEYN 1975, van UDEN 1987).

When freesias are planted in May, June or July the temperature in the first 6—10 weeks is usually high, at least periodically, and the temperature in glasshouses is over 18 °C. To keep the temperature as near optimum as possible the glasshouse is well ventilated and shaded while the soil is covered with some isolating material; sprinklers are often used, too. In spite of these measures the temperature in most summers is too high, and for this reason the timing of the autumn crop has been very

difficult. Soil cooling with cold water allows greater certainty for the flowering of freesias in autumn. This method has been developed in the Netherlands since the 1960s. The soil is cooled by circulating cold water in pipes, which are placed either 30—40 cm deep in the soil or placed on top of the soil (DIJKHUIZEN and van HOLSTEYN 1975, van HARTEN 1986). The water can be cooled by a machine with capacity of about 20—40 W/m², but it is also possible to manage without a cooling machine if there is enough well water available for cooling (DIJKHUIZEN and van HOLSTEYN 1975, van de WIEL-van SON 1984, RAVEN 1988,

RISTIMÄKI 1989). The soil is usually covered with some isolating material, e.g. styrox or straw (van HARTEN 1986, van de WIEL-van SON 1984, van LEEUWEN 1989).

In this study the soil cooling method was applied under Finnish climatic conditions at the Institute of Horticulture in the Agricultural Research Centre of Finland during 1985—88. The aim was to determine the optimum duration for cooling and the optimal combination of extent of cooling and use of covers, and to identify which varieties are best suitable for autumn flowering when using the soil cooling method in northern conditions.

MATERIAL AND METHODS

The soil cooling was carried out at the Institute of Horticulture by circulating cold water in plastic pipes which were placed about 15 cm deep in peat about 25 cm apart from each other. The diameter of each pipe was 32 mm, and there were four pipes, making two cycles in the bed. Each cycle could be disconnected from the circulation so that it was possible to compare beds without cooling to beds with one and two cooling cycles. The water was cooled by a cooling machine with a capacity of 40 W/m² and the temperature of the circulating water was lowered to 8—10 °C. Half of the beds were covered with 2—3 cm styrox grains, and the other half were uncovered. Thus all combina-

tions of covered and uncovered beds with no, one or two cooling cycles could be compared.

Six varieties of normal size corms (5 + , 150—200 corms/kg) were planted at a density of 120 corms/net-m² and about 7 cm deep in order to escape the heating effect of the sun on the surface of the peat. The duration of cooling was 6, 8, 10 or 12 weeks and the cooling started a few days after planting. Some beds were cultivated for the first two weeks without cooling, followed by cooling for 8 weeks. The varieties and trial arrangements are given in Table 1. The flowers were harvested when the first flower of the spike opened. The aim in harvesting was to obtain separate lateral stems with the main

Table 1. The trial arrangements at the Institute of Horticulture in 1985—1988. Symbols: s covered with styrox grains; p peat without cover. 0 without cooling; 1 with one cooling cycle (2 pipes); 2 with 2 cooling cycles (4 pipes). The duration of cooling has 6, 8, 10 or 12 weeks; 2 + 8 weeks = first two weeks without cooling, thereafter cooling for 8 weeks. x marks the presence of control. The varieties were Athene, Aurora, Blue Navy, Oberon, Rosalinde, Royal Blue.

Year	Planting date	Duration of cooling (weeks) in different combinations of cooling cycles and cover					
		0p	0s	1p	1s	2p	2s
85	11/6	x	x	—	10	10	10
86	10/6	x	x	10	10	10	10
87	10/6	x	x	8,12	8,12	6,8,12	6,8,12
88	8/6	x	x	8,10	8,10	8,10,2 + 8	8,10,2 + 8

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stem at least in quality class I. The crop was classified according to the following criteria: in the Extra class the stem height should be at least 45 cm and the flower count at least 7; in class I, correspondingly 35 cm and 5 flowers; in the class II, 30 cm and 4 flowers.

The temperatures in the peat at the depth of the corm were measured daily at 14.00 hours (2 p.m.).

RESULTS

Temperatures

The weather conditions varied very widely during the trial period 1985—1988. June and July of the first two years were near the mean for the period 1931—60 (Table 2), summer 1986 being warmer than summer 1985. The mean temperature of August was near the mean in 1985, but more than 2 degrees below the mean in 1986. The temperatures in June and July are important from the point of view of this study. The summer of 1987 was exceptionally cold: the mean temperatures of the summer months were about 2 degrees below normal. The summer of 1988, on the other hand, was the warmest of all and the mean temperatures of June and July were 2.0—3.2 degrees higher than the mean for 1931—60.

The temperatures in the beds with two cooling cycles and styrox grain cover stayed nearly constant in spite of fluctuations in the outside temperature, and remained below 18 °C except for one or two small deviations (Fig. 1). The temperatures in the beds with one cooling cycle and styrox cover were about one degree higher in 1987 and 2—3 degrees higher in 1988 than in beds with two cooling cycles and in both summers 18 °C or higher. In control beds without cooling and cover the temperature was over 20 °C in June and July of 1988 and 20 °C or a little below in 1987.

Table 2. Mean temperatures °C of June, July and August in 1985—88 and for 1931—60 in Piikkiö, at the Institute of Horticulture.

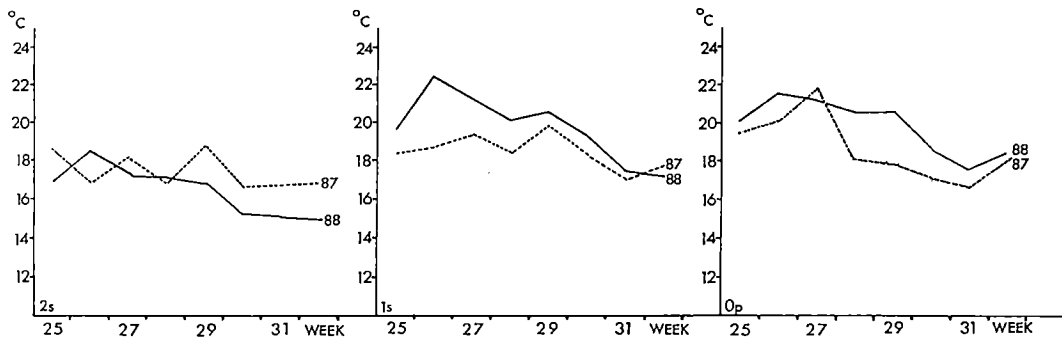| Year	June	July	August
31—60	14.0	17.3	15.9
85	13.8	15.8	16.4
86	16.5	16.4	13.7
87	11.9	15.3	12.2
88	17.2	19.3	14.8

Fig. 1. The soil temperature at the depth of the corm during weeks 25—32 in 1987 – – – and 1988 ——— in the control bed (uncooled and uncovered 0p), in the bed with 1 cooling cycle and styrox cover (1s) and in the bed with 2 cooling cycles, and styrox cover (2s). Temperatures were measured daily at 14:00 hrs.

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Timing of the crop

Every summer the flowering began first in the beds with two cooling cycles and styrox cover (Table 3). The variety 'Rosalinde' was the first to flower, at 13—14 weeks after planting every summer. 'Aurora' required 12—14 weeks to flower, 'Royal Blue' 13—17 weeks, 'Athene' 14—15 weeks, 'Blue Navy' 15—17 weeks and 'Oberon' 16—17 weeks. If no styrox cover was used, the flowering was delayed in 1987 by 1 week, in 1985—86 by 2—3 weeks and in 1988 by 4—8 weeks, depending on the variety.

In the beds with one cooling cycle and styrox the flowering of 'Rosalinde' and 'Aurora' started 13—16 weeks after planting, 'Athene' took 14—16 weeks and 'Royal Blue' 14—19 weeks. 'Oberon' required 16—20 weeks for flowering and 'Blue Navy' 14—21 weeks. The non-use of styrox cover caused a delay of 1—2 weeks in a cold summer and a delay of 2—5 weeks in a hot summer. In all cooled and styrox-covered beds every variety took the longest time from planting to flowering in 1988 and the shortest in 1987.

In the control bed without cooling and cover, too 'Rosalinde' was the first to flower, at 18—21 weeks after planting. 'Aurora' took 20—24 weeks, 'Royal Blue' 20—25 weeks, 'Athene' 23—26 weeks, 'Oberon' 23 weeks and 'Blue Navy' 22—27 weeks or even longer. The styrox cover did not have a clear effect in this bed: in some cases the time required for flowering was shortened by 1—2 weeks with styrox cover, but in other cases the effect was reversed.

The duration of cooling had no noticeable effect on the start of flowering in the beds with two cooling cycles. In the beds with one cooling cycle a longer duration of 10 weeks cooling instead of eight weeks gave an earlier flowering by about 1 week in 1988 with 'Blue Navy', 'Oberon' and 'Rosalinde'. The combination of two weeks without cooling and 8 weeks cooling with 2 cycles delayed flowering by 1—3 weeks.

Table 3. The time from planting to flowering in weeks in 1985—88. Dates of planting: 11.6.1985; 10.6.1986; 10.6.1987; 8.6.1988. Planting density 120 corms/net-m². Symbols: 0 uncooled; 1 cooled with one cycle; 2 cooled with two cycles; p uncovered (peat); s covered with styrox grains 2—3 cm; — no flowering. Cooling times 6, 8, 10 or 12 weeks and 2 + 8 weeks (2 weeks without cooling and 8 weeks cooling). Athene is missing in 1988 and Oberon in 1987.

	Athene			Aurora				Blue Navy				Oberon			Rosalinde				Royal Blue			
	85	86	87	85	86	87	88	85	86	87	88	85	86	88	85	86	87	88	85	86	87	88
0p	26	26	23	21	22	20	24	27	26	22	—	23	23	23	18	20	19	21	20	25	21	24
0s	22	26	23	21	23	19	23	27	—	26	23	21	23	22	18	18	18	21	22	24	24	26
1p 8w			16			13	21			16	23			22			19				17	20
1s 8w			14			13	15			14	21			20			16				14	18
1p 10w		18				20				21	21		17	21		14	18					21
1s 10w	16	15		14	16	14		17	18		19	19	16	17	17	14	16		15	17		19
1p 12w			16			13				16							13				16	
1s 12w			14			13				16							13				14	
2p 6w			16			13				16											17	
2s 6w			14			12				15											13	
2p 8w			14			13	18			16	23			19			18				16	19
2s 8w			14			13	14			15	15			16			13	14			14	17
2p 10w	16	16		17	17	19		19	22		22	17	17	21	14	14	19		15	20		20
2s 10w	15	15		16	15	14		16	17	17	17	14	16	17	13	13	14	13	14	17		16
2p 12w			15			13				16							14				14	
2s 12w			14			13				15							13				14	
2p 2 + 8w							20				22			20				18				21
2s 2 + 8w							16				18			17				17				18

Table 4. Some flower properties measured from flowers grown in uncooled, uncovered beds (0p) and in cooled beds with two cooling cycles and styrox cover (2s) in 1988, showing the distribution of the crop by quality classes.

Variety	Stem height cm		Number of flowers		E%		I%		II%	
	0p	2s	0p	2s	0p	2s	0p	2s	0p	2s
Aurora	44.1	39.9	10.5	9.2	24	2	63	71	9	18
Blue Navy	—	43.3	—	7.6	—	6	—	63	—	25
Oberon	43.4	41.4	9.9	10.0	13	2	70	63	15	25
Rosalinde	40.4	39.4	8.7	9.2	1	—	46	53	27	30
Royal Blue	—	39.4	—	7.2	—	—	—	48	—	22

The flower properties

The soil cooling had little or no effect on the flower properties or quality (Table 4). The height of the stem was decreased by 1.0—4.2 cm by soil cooling, however and since the allocation to quality classes is mainly based on the length of the stem, for this reason there were more Extra class flowers in the uncooled beds.

DISCUSSION

The soil cooling method developed in the Netherlands is very suitable for application to Finnish conditions. In this trial the depth of the soil was 25 cm, and a distance between the pipes of 25 cm and the placing of the pipes 15 cm deep in the peat was enough to keep the temperature even near the corms. The placing of the pipes 40 cm deep, which is recommended in other papers (DIJKHUIZEN and van HOLSTEYN 1975, van de WIEL-van SON 1984) is based on HEYNA (1967), who suggested that the most ideal situation is realized when the distance between the pipes is equal to the depth, and stated that this distance is usually 40 cm. Deeper planting has been recommended in summer (van de WIEL-van SON 1984). A temperature of about 15 °C during flower induction is enough to make the autumn crop sure and to guarantee the quality, however, as was shown in this trial and as is supported by other investigations (van de WIEL and BOEIJE 1988). This temperature was maintained when the water circulating in the pipes was at about 8—10 °C. This temperature has also proved to be optimal for soil cooling in experiments in the USA (DREESEN and LANGHANS 1987). Even if the temperature is as high as 11 °C, however, the cooling effect may be adequate (van de WIEL-van SON 1984).

In this trial the combination of two cooling cycles/bed and styrox grain cover proved to be the most effective in advancing the autumn flowering of freesias. With this method, the flowering starts at least four weeks and even two months earlier than when cultivated traditionally. The extent of the benefit depends on the variety and the weather conditions. Similar results have been obtained by DREESEN and LANGHANS (1987); according to their trial, the combination of one cooling cycle per bed and styrox grain cover can also be useful in timing the autumn flowering, but the benefit is not as clear as with the two-cycle system. Neither cooling nor covering alone has the desired effect. The duration of the cooling, however, did not have a marked effect on the start of flowering. On the basis of this trial, if the temperature in the peat is near the optimum, about

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15 °C, a cooling period of six weeks is recommended. There is no harm in a longer duration, but it usually gives no added advantage. If the temperature is just a little over the optimum but not over 18 °C, 8—10 weeks is recommended. The best time to start cooling is some days after planting.

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SELOSTUS

Kasvualustan jäähdytyksen vaikutus kesällä istutetun freesian kukintaan

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Maatalouden tutkimuskeskus

Freesian kukintoaiheen kehittyminen on voimakkaasti lämpötilasta riippuvainen. Erityisesti 6—10 ensimmäisen viikon aikana istutuksen jälkeen kasvualustan lämpötila ei saisi ylittää 18 °C, ja edullisin lämpötila on 13—15 °C. Syyskukintaa varten freesiat istutetaan toukokuun lopussa tai kesäkuun alussa, jolloin kasvihuoneen lämpötila nousee usein kukintoaiheen kehittymisen aikaan yli 18 °C:een. Etenkin hyvin lämpiminä kesinä kukinta viivästyy tai estyy kokonaan, ellei lämpötilan säätelyyn ole käytettävissä muita keinoja kuin tuuletus, varjostus ja sumutus. Kierrättämällä kasvualustaan upotetuissa putkissa kylmää vettä ja kattamalla turpeen pinta styroksirouheella tutkimuksissa saatiin kukinnan ajoitus varmaksi. Jäähdytysveden lämpötila oli 8—10 °C, ja sen avulla kasvualustan lämpötila pysyi 13—16 °C:ssa, jos putkikiertoja upotettiin turpeeseen 2 kpl ja pinta peitettiin 2—3 cm:n styroksirouhekerroksella. Jos jäähdytyskiertoja oli penkissä 1 kpl, lämpötila pysyi 1—3 °C korkeampana. Aikaisimman lajikkeen kukinta aikaistui 5—7 viikkoa kahta jäähdytyskiertoa ja styroksikatetta käytettäessä, myöhäisimmän lajikkeen vastaavasti 8—10 viikkoa. Tulos oli tehokkaimmin jäähdytetyissä penkeissä jokseenkin sama koekesien hyvin erilaisesta säästä huolimatta. Riittävä jäähdytysaika oli 6—10 viikkoa jäähdytyksen tehokkuudesta riippuen.

ROOTING *IN VITRO* OF MICROPROPAGATED BARBERRY (*BERBERIS THUNBERGII*) SHOOTS

SAILA KARHU and KAIJA HAKALA

KARHU, S. and HAKALA, K. 1990. Rooting *in vitro* of micropropagated barberry (*Berberis thunbergii*) shoots. Ann. Agric. Fenn. 29: 179—185. (Agric. Res. Centre, Inst. Hort., SF-21500 Piikkiö, Finland.)

Berberis thunbergii cuttings prepared from shoots proliferated *in vitro* rooted readily when rooting was induced for 7 days in the dark in a liquid medium containing 58.4 mM saccharose and 2—6 µM IAA or IBA, and the shoots transferred thereafter into auxin-free agar-solidified medium with half-strength Murashige and Skoog (MS) salts. There was no significant difference in rooting response after IAA and IBA treatments at equal molarities, the rooting percentage being 70—95 % after all auxin treatments, while the percentage rooting was only 25 % without auxin supplement. The length and number of roots was also increased significantly by all auxin treatments. Transfer of shoots into IAA-supplemented (4 µM) rooting medium after IAA-initiation (4 µM) resulted in significantly poorer rooting percentages and shorter roots than transfer into auxin-free rooting medium. Supplementation of H_3BO_4 (0.15 mmol l^{-1}) and $CaCl_2$ (4.5 mmol l^{-1}) during initiation and rooting had an adverse effect on rooting, when IAA was included in the rooting medium. Addition of both H_3BO_4 and $CaCl_2$ into the root initiation and rooting medium inhibited rooting totally. Callus was present at all shoot bases, rooted or unrooted, when IAA was included in the rooting medium.

Index words: *Berberis thunbergii*, micropropagation, rooting, auxins, boron, calcium.

INTRODUCTION

Barberry, especially *Berberis thunbergii*, is a popular woody ornamental in Finland. Barberry plants propagated by seed are frequently used for hedges and mass plantings. The variable overwintering of barberry plants has given rise to demand for a uniform, winterhardy plant material. A procedure for micropropagation of *B. thunbergii* clones is therefore being investigated in our laboratory.

Rooting is known to be the critical step in the micropropagation of a number of woody species, with both physical and chemical factors playing important roles (WILKINS and

DODDS 1983, GEORGE and SHERRINGTON 1984, ZIMMERMAN and FORDHAM 1985, BONGA and DURZAN 1987). UNO and PREECE (1987) reported poor root formation and inefficiency of auxin treatments on rooting of *B. thunbergii atropurpurea*. According to our preliminary observations, dark treatment during the initiative phase of rooting has proved to be useful for rooting of *B. thunbergii*. This has also been reported on some other species (DRUART et al. 1982, ZIMMERMAN and FORDHAM 1985, NORTON and NORTON 1988).

Interaction of boron and calcium with aux-

ins and their necessity for root development has been frequently demonstrated (BURSTRÖM 1952, JARVIS et al. 1983, 1984, HAISSIG 1986, MARSCHNER 1986). The promotive effect of boron-calcium interaction for root primordia formation and later root development has also been shown (MIDDLETON et al. 1978). Both boron and calcium are considered to be immobile elements in plants (MARSCHNER 1986), which explains the necessity of exogenous supplement of these nutrients for root formation (HAISSIG 1986).

This paper presents experiments made to investigate rooting responses of micropropagated barberry shoots to supplements of auxins, calcium and boron in the root initiation phase in the dark and during the subsequent rooting phase.

MATERIAL AND METHODS

Microculture of *B. thunbergii* was initiated by axillary bud culture. Microshoots were propagated by axillary branching on a medium containing MURASHIGE and SKOOG (1962) (MS) salts supplemented with 0.56 mM myo-inositol, 4.1 µM nicotinic acid, 2.4 µM pyridoxine-HCl, 1.2 µM thiamine-HCl, 26.6 µM glycine, 8.9 µM benzyladenine (BA), 1.4 µM gibberellic acid (GA_3), 58.4 mM saccharose and 7 g l^{-1} of Difco Bacto-agar. Iron was supplied as ethylenediaminetetraacetic acid, iron (III) -sodium salt, 36.7 mg l^{-1}. The pH was adjusted to 5.7 before autoclaving.

For shoot proliferation, cultures were grown at 25 ° ±2 °C with 16 h photoperiods provided by mixed light (Airam warm white de luxe 40 W + Airam floralux 40 W) at a photon flux density of 35 µmol $m^{-2}s^{-1}$. The same conditions were used for rooting in light. Dark treatments were performed by wrapping root initiation vials in aluminium foil and placing them in the growth room.

For each root initiation treatment, the proximal 1 cm of 20 healthy shoots about 2 cm long with leaves retained only at the distal 1 cm of the cutting were placed into initiation medium (modified from ZIMMERMAN and FORDHAM 1985) containing 58.4 mM saccharose + auxin and mineral supplements according to each experiment. pH was adjusted to 5.4 before autoclaving. After 7 days in darkness, the shoots were brought into the light and transferred into solid medium (Difco Bacto-agar 0.7 %) otherwise similar to the propagation medium above, but containing 50 % of MS salts, no BA or GA_3 supplemented and pH adjusted to 5.6. Auxins and extra minerals were supplemented according to each experiment.

Experiment A. All the 5 initiation media contained 4 µmol l^{-1} of indoleacetic acid (IAA). One medium was also supplemented with 0.15 mmol l^{-1} of H_3BO_4, one with 4.5 mmol l^{-1} of $CaCl_2$ and one with both 0.15 mmol l^{-1} of H_3BO_4 and 4.5 mmol l^{-1} of $CaCl_2$. The subsequent 5 rooting media contained the same hormone and mineral supplements as the initiation media, except for one medium which was auxin free.

Experiment B. Of the 7 root initiation media, one was auxin free, three contained IAA in amounts of 2, 4 and 6 µmol l^{-1} and three contained indolebutyric acid (IBA) in amounts of 2, 4 and 6 µmol l^{-1}. The rooting medium after all initiation treatments was auxin free.

Rooting was observed 1 month after transfer of the shoots to solid media.

All data on root number and length in experiment B were subjected to a one-way analysis of variance performed by the SAS general linear models procedure (GLM) (SAS® User's Guide: Statistics, Version 5 Edition, 1985). Contrasts of treatments were also tested by GLM. In ex-

periment A, rooting was too poor to be analyzed by analysis of variance. In both experiment A and B, differences in percentage rooting between treatments were evaluated by performing Chi-square analysis on the number of cuttings rooted in each treatment. To calculate χ^2, the correction for continuity (Yates) was made.

<center>RESULTS</center>

Experiment A

Rooting was best when the shoots were transferred from initiation medium to auxin free rooting medium (treatment 1). In this medium 85 % of the shoots rooted, which was significantly more (p = 0.01) than in treatment 2 (rooting medium with 4 μmol l^{-1} IAA) (Table 1). The mean number of the roots in rooted shoots was about 3 and the mean length of the roots was about 12 mm, which was many times greater than in other treatments (Table 2).

Added extra minerals had an adverse effect on rooting when IAA was included in the rooting medium. In treatments 4 and 5 the rooting percentage was significantly poorer than in treatment 2 (at the level of p = 0.05). Addition of both $CaCl_2$ and H_3BO_4 simultaneously inhibited rooting totally (Table 1). The roots were very short in all rooting media containing IAA. Both rooted and non-rooted shoots in treatments 2—5 had brown-coloured callus at their bases (Table 2).

Experiment B

All auxin treatments during the root initiation phase improved the rooting percentage significantly (treatment 1 (no auxin) vs. treatments 2 and 7 at the level of p = 0.001, treatment 1 vs. treatments 3, 4, 5 and 6 at the level of p = 0.01) (Fig. 1). The length of roots in rooted shoots was increased by all the auxin treatments (F = 16.88, p = 0.0001), the mean length of roots in rooted shoots being about 3—4 times greater after auxin induction as compared to treatment

Table 1. Rooting percentage of micropropagated *B. thunbergii* shoots in rooting experiment A. Mean values followed by the same letter are not significantly different at the 5 % level (chi-square test). Twenty cuttings took part in each treatment.

Treat-ment	Initiation medium		Rooting medium	Rooting %
1.	IAA 4 μM	$\longrightarrow$	auxin free	85 A
2.	IAA 4 μM	$\longrightarrow$	IAA 4 μM	35 B
3.	IAA 4 μM + Ca	$\longrightarrow$	IAA 4 μM + Ca	15 CB
4.	IAA 4 μM + B	$\longrightarrow$	IAA 4 μM + B	5 C
5.	IAA 4 μM + Ca + B	$\longrightarrow$	IAA 4 μM + Ca + B	0 C

Table 2. Number of rooted shoots (N) of micropropagated *B. thunbergii* and mean number ($\pm$SE) and mean length ($\pm$SE) of roots in rooted shoots in experiment A. Presence of callus (C) at shoot bases indicated by YES (callus present) and NO (callus absent). Twenty cuttings took part in each treatment.

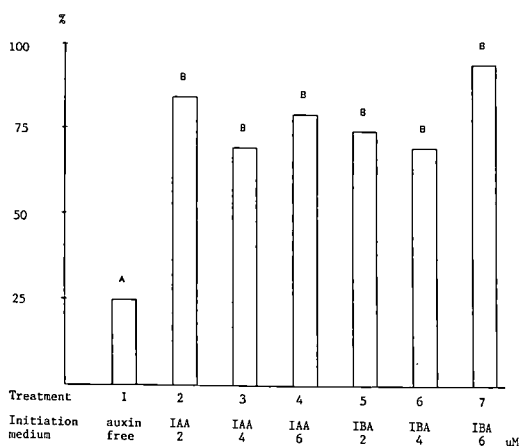| Treat-ment	Initiation medium		Rooting medium	N	Mean number $\pm$SE	Mean length (mm) $\pm$SE	C
1.	IAA 4 μM	$\longrightarrow$	auxin free	17	3.35 $\pm$ 0.44	11.62 $\pm$ 1.89	NO
2.	IAA 4 μM	$\longrightarrow$	IAA 4 μM	7	2.29 $\pm$ 0.44	2.14 $\pm$ 0.40	YES
3.	IAA 4 μM + Ca	$\longrightarrow$	IAA 4 μM + Ca	3	3.67 $\pm$ 0.72	2.25 $\pm$ 0.12	YES
4.	IAA 4 μM + B	$\longrightarrow$	IAA 4 μM + B	1	1.00 —	2.00 —	YES
5.	IAA 4 μM + B + Ca	$\longrightarrow$	IAA 4 μM + B + Ca	0	— —	— —	YES

Fig. 1. Rooting percentage of micropropagated B. thunbergii shoots in rooting experiment B. Mean values followed by the same letter (on top of each bar) are not significantly different at p = 0.05 (chi-square test). Twenty cuttings took part in each treatment.

Table 3. Number of rooted shoots (N) of micropropagated B. thunbergii and mean number (±SE) and mean length (±SE) of roots in rooted shoots in experiment B. There was no callus at the shoot bases irrespective of the treatment. Twenty cuttings took part in each treatment.

Treatment	Initiation medium	N	Mean number ±SE	Mean length (mm)±SE
1.	auxin free	5	1.60±0.24*	3.60±1.31***
2.	IAA 2 µM	17	2.70±0.34	12.97±1.80
3.	IAA 4 µM	14	4.00±0.58	10.14±1.41
4.	IAA 6 µM	16	4.00±0.62	13.07±2.10
5.	IBA 2 µM	15	2.47±0.36	13.52±1.88
6.	IBA 4 µM	14	2.64±0.58	13.93±1.66
7.	IBA 6 µM	19	5.11±0.88	10.58±1.25

* = treatment differs from other treatments at the level of p = 0.05.
*** = treatment differs from other treatments at the level of p = 0.001.
(SAS general linear models procedure, contrast test)

1 (Table 3). Between the different auxin treatments, on the other hand, root length did not differ significantly. The number of roots was also significantly increased by all auxin treatments as compared to treatment 1 (F = 4.02, p = 0.0478), with no significant difference between the IAA and IBA treatments. The highest number of roots in rooted shoots was achieved by root initiation medium supplemented with 6 µmol l^{-1} of IBA (Table 3), the effect of IBA supplement being significantly linear (F = 8.29, p = 0.0050) (GLM contrast test) (Table 3). No callus was found at the bases of the shoots.

DISCUSSION

Shoots of several woody ornamental and fruit species can be rooted in vitro in the continued presence of exogenous auxin (e.g. WILKINS and DODDS 1983, MEZZETTI et al. 1988, NORTON and NORTON 1988). However, the presence of supplied auxin can also disturb the organization of the root primordia and their subsequent growth and lead to excessive callusing at the bases of shoots (JAMES and THURBON 1979, JARVIS et al. 1983, COLLET 1988). The experiments reported here show that B. thunbergii belongs to the latter group of plants: supplied auxin is beneficial in the root initiation phase, but inhibits the development of adventitious roots in a later phase, leading to the formation of brownish shoot base callus.

The results of experiment A suggest that rooting is not promoted by incorporating boron in the root initiation medium and adding the same amount of it to the rooting medium, although the essential role of boron supplement in the development of adventitious roots in stem cuttings has been frequently reported (HAISSIG 1986, JARVIS 1986, MARSCHNER 1986). Further, high concentrations of supplied auxin have been shown to enhance rooting of cuttings, especially when relatively high concentrations of boron are also supplied. The role of boron in root formation has been proposed to be one of controlling effective auxin concentrations through its effect on IAA oxidase activity (BOHNSACK and ALBERT 1977, JARVIS 1986).

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Boron would thus act by diminishing the level of auxin that initiates rooting, but would be inhibitory to subsequent root organization and growth. In the present experiment (A) this kind of operation was not found: callus also formed at the bases of shoots incubated in the presence of both IAA and boron, which indicates no diminished level of excess auxin during the root growth phase. The effect of boron, in fact, seemed to be adverse: boron supplement resulted in significantly lower rooting percentages compared to shoots without boron supplement, although no symptoms of boron toxicity were seen in the shoots.

This result might be explained by the fact that only one concentration of boron and one of IAA was tested, the rooting response being related to the amounts of both auxin and boron (JARVIS et al. 1984). On the other hand, it has also been suggested that the primary role of boron in plants might be concerned with nucleic acid metabolism and the control of cytokinesis (ALI and JARVIS 1988), or might operate through effects on auxin transport. Although not related to the acropetal movement of exogenous auxin, both boron and calcium availability have been shown to be essential to the basipetal movement of IAA in sunflower hypocotyl segments (TANG and DELA FUENTE 1986 a, 1986 b).

Although we did not measure the polar transport of auxin, the poor rooting of shoots supplied with boron might be explained by the auxin transport model. Callus growth at the bases of shoots indicates a supraoptimal level of auxin in the root organization and elongation phase (experiment A, treatments 2—5), as mentioned before. If the model of boron-promoted enhancement of basipetal transport of endogenous IAA holds true, very high auxin levels must have arisen at the shoot bases in treatments 4 and 5, affecting rooting adversely.

Similarly to boron, calcium has also been shown to be essential to basipetal auxin transport and to the mechanism of auxin action in higher plants (DELA FUENTE and LEOPOLD 1973, HERTEL 1983, TANG and DELA FUENTE 1986 a, 1986 b). The results of experiment A indicate that in the continuous presence of exogenous IAA, calcium addition does not significantly affect rooting of *Berberis* microshoots. This might be explained by the fact that the effect of calcium on root formation is most frequently connected with the elongation phase of roots (HAISSIG 1986, HASENSTEIN and EVANS 1986), and all the rooting media contained at least 1.5 mM $CaCl_2$. Further, in contrast to the effect of boron deficiency on auxin transport, the effect of calcium deficiency on IAA transport in stem segments can be completely reversed with subsequent calcium supplement (TANG and DELA FUENTE 1986 b). Due to this, normal auxin action could be attained in shoots during the shoot elongation phase, even if the root initiation medium was calcium-free.

The results of experiment B confirm that good root formation and development is attained in an auxin-free medium if shoots of *B. thunbergii* are supplied with auxin during the preceding root initiation phase. Even without any auxin treatment, one cutting in four rooted, but the roots remained short. This rooting can be presumed to result from endogenous auxin.

Both the rooting percentage and root length were satisfactory no matter which of the tested IAA or IBA concentrations was used. The highest number of roots was formed when the highest amount of IBA was supplied in the initiation phase of rooting. The higher efficiency of IBA in relation to the equal molarities of IAA, which has been reported earlier (MIDDLETON et al. 1978, JARVIS 1986) was, however, not noted in this study.

Incubation of microshoots for 7 days in darkness was found to be advantageous for rooting of *Berberis* in our preliminary observations. Darkness in the initiation phase of root formation has also produced good results in *Malus* (DRUART et al. 1982, ZIMMERMAN and FORDHAM

1985), *Prunus* and *Spiraea* (NORTON and NORTON 1988) and *Eucalyptus* (DAS and MITRA 1990). Furthermore, rooting cuttings of *Berberis in vivo* has been reported to be more successful after etiolating the stock plants in the dark (KNOX and HAMILTON 1982); this was not the case, however, in another experiment in our laboratory.

Lack of dark treatment may have been the reason for the earlier limited success in the rooting of micropropagated *Berberis* shoots (UNO and PREECE 1987). On the other hand, one must not forget the differences in rooting response between different cultivars and genotypes, a fact well known by every plant propagator.

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SELOSTUS

Japaninhappomarjan (*Berberis thunbergii*) mikrolisättyjen versojen juurrutus *in vitro*

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Maatalouden tutkimuskeskus

Mikrolisätyn japaninhappomarjan (*B. thunbergii*) juurtumis-
ta tutkittiin kahdella kaksivaiheisella kokeella. Ensimmäi-
sessä vaiheessa (induktiovaihe) n. 2 cm:n mittaisia verson-
kärkiä pidettiin 7 vrk:n ajan pimeässä, versojen tyvet 1 cm:n
matkalta liuoksessa, joka koostui vedestä, sokerista (58,4
mM sakkaroosi), kokeiltavista auksiineista sekä lisämineraa-
leista, koejäsenestä riippuen. Toisessa vaiheessa (juurrutus-
vaihe) versot siirrettiin valoon, kiinteälle alustalle (0,7 %
Difco Bacto-agar), johon oli lisätty 50 % Murashigen ja
Skoogin alustan mineraaleista, 58,4 mmol l⁻¹ sakkaroosia,
0,56 mmol l⁻¹ myo-inositolia, 4,1 μmol l⁻¹ nikotiinihap-
poa, 2,4 μmol l⁻¹ pyridoksiini-HCl:a, 1,2 μmol l⁻¹ thia-
miini-HCl:a ja 26,6 μmol l⁻¹ glysiiniä sekä auksiineja ja li-
sämineraaleja koejäsenestä riippuen.

Kokeessa A jokaisen 5 koejäsenen induktioalustaan oli
lisätty 4 μmol l⁻¹ indolietikkahappoa (IAA). Yhteen koejä-
seneen oli lisätty myös 4,5 mmol l⁻¹ CaCl₂:a, yhteen 0,15
mmol l⁻¹ H₃BO₄:ää sekä yhteen molempia mineraaleja
e.m. määrät. Juurrutusalustoissa olivat samat lisäykset, paitsi
yhdessä koejäsenessä, jossa juurrutusalusta oli auksiiniton
ja ilman lisämineraaleja. Kokeessa B juurrutusalusta oli kai-
kissa koejäsenissä auksiiniton. Induktioalustaan oli lisätty
joko IAA:ta tai indolivoihappoa (IBA) 2, 4 tai 6 μmol l⁻¹.
Kontrollina oli koejäsen, jonka induktioalustassa ei ollut auk-
siinilisäystä.

Kokeessa A juurrutus onnistui hyvin vain koejäsenessä,
joka IAA-induktion jälkeen siirrettiin auksiinittomalle juur-
rutusalustalle (juurtumisprosentti oli 85 %). Tällä alustalla
juuret olivat myös huomattavasti pidempiä (keskimäärin n.
12 mm) kuin auksiinipitoisella alustalla. Auksiinipitoisella
alustalla juurtuminen oli heikkoa. Mineraalilisäykset vähen-
sivät juurtumista: versoista, joiden juurtumisinduktio- ja
juurrutusalustaan oli lisätty ylimääräistä CaCl₂:ää, vain
15 % juurtui. Kun juurtumisinduktio- ja juurrutusalustaan
oli lisätty ylimääräistä H₃BO₄:a, vain yksi verso 20:stä juur-
tui. Molempien mineraalien samanaikainen lisäys johti juur-
tumisen täydelliseen estymiseen. Kallusta oli sekä juurtu-
neiden että juurtumattomien versojen tyvellä, kun juurru-
tusalustassa oli auksiinia.

Kokeessa B kaikki auksiinikäsittelyt johtivat hyvään juur-
tumiseen (70—95 % versoista juurtui). IAA:n ja IBA:n vai-
kutuksilla ei ollut merkitsevää eroa. Auksiinittomalla induk-
tioalustalla olleista versoista vain 25 % juurtui. Juurten pi-
tuus ja lukumäärä lisääntyivät kaikkien auksiinikäsittelyjen
vaikutuksesta.

Mikrolisätyn japaninhappomarjan versot juurtuvat siis hy-
vin, kun juurtumista ensin indusoidaan n. yhden viikon ajan
pimeässä, induktioliuoksessa, johon on lisätty joko IAA:ta
tai IBA:ta 2—6 μmol l⁻¹ ja versot siirretään sen jälkeen va-
loon, auksiinittomalle agar-alustalle.

EFFECT OF *STREPTOMYCES* SP. ON SEED-BORNE FOOT ROT DISEASES OF WHEAT AND BARLEY

I. POT EXPERIMENTS

RISTO TAHVONEN and HANNA AVIKAINEN

TAHVONEN, R. & AVIKAINEN, H. 1990. Effect of *Streptomyces* sp. on seed-borne foot rot diseases of wheat and barley. I. Pot experiments. Ann. Agric. Fenn. 29: 187—194. (Agric. Res. Centre, Inst. Pl. Protect., SF-31600 Jokioinen, Finland.)

A powdery, biological control preparation, containing *Streptomyces griseovirides* as the biocontrol agent, was tested against foot rot diseases of wheat and barley in the form of seed treatment in pot experiments. A dosage of 3—15 g powdery preparation/kg seed decreased damage caused by *Fusarium* spp. and *Bipolaris sorokiniana* on both inoculated and uninoculated seed. A mercurial seed treatment gave better control than *Streptomyces*. The dry weight of the shoots was higher after *Streptomyces* treatment than in untreated plants. The seed treatment decreased the contents of *Fusarium* spp. and *B. sorokiniana* when the seeds were tested by the seed contamination method. The effect of seed treatment remained stable for 2—3 weeks when the treated seeds were stored in dry conditions.

Index words: wheat, barley, biological control, *Streptomyces griseovirides*, *Fusarium culmorum*, *Bipolaris sorokiniana*.

INTRODUCTION

Fusarium culmorum and *Bipolaris sorokiniana* are common pathogens on barley and wheat under Finnish conditions (MÄKELÄ and PARIKKA 1980, KURPPA 1984). These pathogens are usually controlled by dusting the seed with a mercury preparation (KURTTO et al. 1988). However, mercury dusting is gradually being phased out of commercial use in Finland, and mercury-free preparations are being sought to replace it (KURTTO 1988). In addition to chemical methods, biological methods, especially bacterial and fungal antagonists, have been tested for seed treatment (BAKER and COOK 1974).

One particular target of biological control is *Gaeumannomyces graminis*, which has been difficult to control by chemical means and which is a serious, world-wide causal agent of foot rot disease on wheat. The most common control targets of chemical dusting are seed-borne pathogens. The first realistic target of biological control would therefore be the control of pathogens, carried on the surface of the seeds, by dusting with antagonists.

The *Streptomyces* bacteria are very common organisms in the soil and are well known for their ability to produce antibiotics. The use of *Streptomyces* sp. isolated from peat in the biological control of plant pathogens has been actively studied in Finland (TAHVONEN 1982). The

main targets of this series of studies have been the control of soil-borne pathogens of greenhouse plants, and seed dusting. Barley and wheat have also been included in the methodological studies and pilot field trials. Seed-borne *F. culmorum* and *B. sorokiniana* fungi have been well or satisfactorily controlled by means of seed treatment in both pot and field experiments (TAHVONEN 1982, 1985, 1988).

The study described in this report is the laboratory and pot experiment part of a more extensive study on the suitability of a powdery preparation of *Streptomyces,* bearing the commercial name of Mycostop, for the biological control of pathogens on barley and wheat in order to replace chemical seed dusting. The aim of the pot experiments was to preliminary test the applicability of this biological control method on cereals, and to obtain information about the correct dusting doses and to locate seed lots with a suitable degree of infection by pathogens for field experiments. The experiments were carried out during 1985—1987 at the Department of Plant Pathology of the Agricultural Research Centre of Finland.

MATERIAL AND METHODS

The seeds were dusted with 3—15 g of a powdery microbial preparation of *Streptomyces griseovirides*/kg seed (TAHVONEN and AVIKAINEN 1987). The preparation contained 10^8—10^9 cfu/g. The dusting doses used in the different experiments are given in the tables. The controls and comparison consisted of non-dusted seed and chemical dusting with alkoxyalkylmercury ''Ceresan'' at a dosage of 2 g/kg seed. Dusting was done by carefully shaking known amounts of seed and dusting agent together in glass conical flasks. In addition to dusting, spraying the surface of the sowing substrate was also tested in 1985. The substrate was sprayed with 1 ml of a 10^{-2}—10^{-4} dilution of *Streptomyces*/pot immediately after sowing before the seeds were covered with soil.

Artificially infected barley seeds were used in three barley pot experiments in 1985. The seeds were washed with an aqueous suspension of *Fusarium culmorum* and *Bipolaris sorokiniana* and then dried for one day at room temperature. The inoculum was prepared by homogenising the 2-week-old fungal mycelium and spores from two PDA petri dishes (Ø = 9 cm) in 500 ml of water. The Pokko barley cultivar was used in the infection experiments.

39 lots of wheat seed and 19 lots of barley seed, naturally infected with *F. culmorum* and *B. sorokiniana,* were tested in pot experiments in 1986—87. The seed lots were obtained from different wholesale seed suppliers on the basis of information supplied by the State Seed Testing Institute. The varieties used are given in Tables 2 and 3. In addition, the seed lots used in the field tests were tested in further experiments in 1986—87. The Pokko variety was also used in these infection experiments.

Retention of the effectiveness of the *Streptomyces* preparation on dusted seed was investigated by sowing infected and dusted seed 14 times at intervals of 0—25 weeks after dusting. The treated seeds were stored at room temperature in paper bags.

Steam-sterilized horticultural peat was used as the substrate. The peat was limed before sterilization at a level of 8 kg dolomitic limestone/m^3 and fertilized following sterilization with peat basic fertilizer. Non-sterilized, limed and fertilized peat was used, in addition to steam-sterilized peat, in the first experiment (Table 1). 3 l plastic pots or 30 × 40 cm plastic boxes were used as the raising containers. There were three or four replicates in each ex-

periment, each replicate consisting of 50 seeds. The growing temperature was 18—20 °C, and illumination about 8 000 lux with multimetal lamps for 16 hours/day. The same illumination and temperature conditions were used in all the experiments.

The degree of infection of the shoots was determined about 3 weeks after sowing using a scale of 0—2, where 0 = healthy, 1 = slightly damaged, 2 = dead or unrecoverable shoot. The dry weight of the shoots was determined by cutting them off at the foot and drying them for 1.5 days at 105 °C. The germination percentage, viability and *F. culmorum* and *B. sorokiniana* contents of the naturally infected seed lots were determined using the blotter method by keeping the seeds for two weeks on moist filter paper in petri dishes ($\varnothing = 14$ cm): 4×25 seeds/treatment and cultivar.

The mean infection indices, dry weights and sprouting-%, *Fusarium*-% and *B. sorokiniana*-%, as determined from the seed tests, are presented in the tables. The dusting effect-%, in which an uninfected seed has the value 100 and an infected one 0, have been calculated from the sowing time experiment. The results have been treated, where necessary, using analysis of variance. Statistical significances are expressed by asterixes (*** > P 0.001, ** > P 0.01, * > P 0.05) and when necessary the calculated LSD at the 0.05 % risk level.

RESULTS

Dusting of artificially infected seed with the powdery preparation of *Streptomyces griseovirides* reduced to a highly significant degree the damage caused by *Fusarium* spp. and *B. sorokiniana* fungi on barley (Tables 1 and 2, Fig. 1). Both chemical and biological control increased the dry weight of the shoots (Table 3). Chemical mercurial dusting gave significantly better control results than *Streptomyces* dusting. There were no differences in the sprout-

Table 1. Effect of *Streptomyces* seed treatment and soil spraying on foot rot of barley caused by *Fusarium culmorum* and *Bipolaris sorokiniana* in fresh (I) and steamed (0) peat.

| | Inoculation of seeds | | | | | |
| | Uninoculated | | F. culmorum | | B. sorokiniana | |
	I	0	I	0	I	0
	Disease index, 0—2					
Untreated	0.08	0.17	0.74	0.70	0.33	0.50
Streptomyces						
5 g/kg seed	0.05	0.10	0.22	0.39	0.04	0.14
10 g/kg seed	0.03	0.07	0.11	—	0.05	0.05
15 g/kg seed	0.02	0.02	0.14	0.28	0.08	0.15
Soil spraying						
10^{-4}	0.08	0.19	0.43	—	0.26	0.57
10^{-3}	0.07	0.21	0.40	0.51	0.26	0.38
10^{-2}	0.03	0.13	0.32	0.46	0.13	0.18
Ceresan 2 g/kg	0.0	0.01	0.01	—	0.01	0.01
F-values:	Seed inoculation		Treatment		Interaction	
Fresh peat (I)	8.93***		34.62***		8.27***	
Steamed peat (0)	48.96***		38.06***		10.68***	

Table 2. Effect of *Streptomyces* seed treatment on foot rot of barley caused by *Bipolaris sorokiniana* and *Fusarium culmorum*.

Treatment	Inoculation of seeds			
	Uninoculated	B. sorokiniana	F. culmorum	Mean
	Disease index, 0—2			
Untreated	0.36	0.20	0.26	0.27
Streptomyces				
5 g/kg	0.07	0.12	0.20	0.13
10 g/kg	0.06	0.12	0.14	0.11
15 g/kg	0.08	0.08	0.13	0.11
Mean	0.14	0.13	0.18	
	Dry weight of shoot			
Untreated	2.73	2.57	2.67	2.66
Streptomyces				
5 g/kg	0.03	2.80	2.83	2.89
10 g/kg	3.23	2.70	2.83	2.92
15 g/kg	3.03	3.07	2.73	2.94
Mean	3.01	2.79	2.77	
F-values	Inoculation	Treatment	Interaction	
Disease index	4.32**	23.65***	4.26**	
Dry weight	3.64**	3.26**	2.33*	

Table 3. Effect of *Streptomyces* seed treatment of foot rot on different cultivars of wheat and barley.

Cultivars and number of lots		Untreated	Streptomyces		Ceresan
			5 g/kg	10 g/kg	
		Disease index, 0—2			
Wheat					
Cv. Luja	16	0.61	0.39	0.39	0.12
Tapio	11	0.84	0.69	0.63	0.17
Other	12	0.48	0.31	0.28	0.11
Mean		0.72	0.56	0.51	0.14
Barley					
Cv. Potra	9	0.81	0.69	0.64	0.17
Arra	3	0.51	0.47	0.37	0.11
Pokko	3	0.43	0.29	0.25	0.07
Eero	2	0.69	0.52	0.51	0.16
Hja 673	2	0.38	0.42	0.26	0.02
Mean		0.56	0.48	0.41	0.11

ing-% between the different dusting treatments.

Streptomyces dusting of uninfected barley and wheat seed which naturally contained varying amounts of *Fusarium* spp. and *B. sorokiniana* reduced foot damage in the sprouting experiments compared to the undusted seed. However, the result was inferior to that obtained with mercury dusting (Table 3). There were no differences in the sprouting-% between the treatments.

Streptomyces dusting reduced the germina-
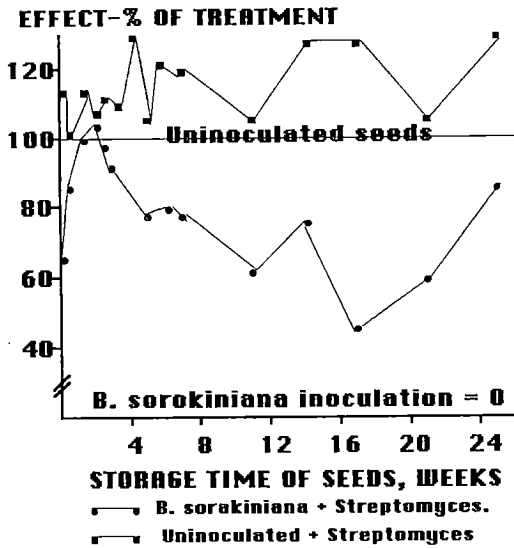
EFFECT-% OF TREATMENT

Fig. 1. Effect of *Streptomyces* seed treatment and storage time of seeds on foot rot of barley caused by *Bipolaris sorokiniana* in a pot experiment.

Table 4. Effect of *Streptomyces* seed treatment on the germination, mortality, *Fusarium* and *Bipolaris* content of wheat and barley seeds as determined by the blotter method.

Cultivars and number of lots		Untreated	*Streptomyces*		Ceresan
			5 g/kg	10 g/kg	
Germination %					
Wheat					
Cv. Luja	16	82	84	83	87
Tapio	11	85	84	84	86
Others	12	72	73	74	78
Mean		80	80	80	84
Mortality %					
Luja	16	13	8	6	2
Tapio	11	9	4	2	2
Others	12	15	9	4	1
Mean		12	7	4	2
Fusarium %					
Luja	16	27	16	13	2
Tapio	11	30	13	12	7
Others	12	37	26	15	7
Mean		31	18	13	5
Germination %					
Barley					
Cv. Potra	9	68	56	45	82
Arra	3	84	67	49	87
Pokko	3	80	74	68	84
Eero	2	57	44	38	88
Hja 673	2	91	82	88	93
Mean		74	62	55	85
Fusarium %					
Potra	9	31	18	12	8
Arra	3	23	11	5	3
Pokko	3	12	6	7	3
Eero	2	14	16	9	0
Hja 673	2	15	13	7	3
Mean		23	15	9	5
Bipolaris %					
Potra	9	5	3	2	0
Arra	3	11	4	5	0
Pokko	3	26	23	19	2
Eero	2	28	24	17	0
Hja 673	2	28	14	15	0
Mean		14	10	8	0

tion of barley in the tests performed by the blotter method, but had no effect on wheat germination. The *Fusarium*-% was lower on the dusted barley and wheat seeds than on undusted ones. Mercury dusting of barley seed completely controlled *B. sorokiniana* but *Streptomyces* dusting afforded only partial control (Table 4).

There were no significant differences in the effectiveness of control given by different dusting doses, but the higher levels (10 g and 15 g/kg) usually gave a slightly better numerical result than 5 g/kg (Tables 1, 2, 3, 4, 5). Spraying the preparation on the substrate reduced foot rot damage on barley, but the result was inferior to that with dusting. There were more cases of foot rot damage on the steam-sterilized peat than the unsterilized peat, but *Streptomyces* dusting decreased the incidence of damage to the same extent on both substrates (Table 1).

The effectiveness of *Streptomyces* dusting was retained for 3—4 weeks on dusted seed stored at room temperature, but subsequently decreased with longer storage periods. Dusting of uninfected seed always resulted in healthier shoots than the control plants right up until the

Table 5. Effect of *Streptomyces* seed treatment on the diseasé index of uninoculated wheat and barley in 1986—1987.

Year and number of seed lots	Untreated	Streptomyces		Ceresan
		3 g/kg	10 g/kg	
		Disease index, 0—2		
Wheat				
1986, 6	0.83	0.80	0.78	0.32
1987, 4	0.33	0.21	0.18	0.04
Barley				
1986, 4	0.80	0.75	0.72	0.21
1987, 3	0.24	0.18	0.13	0.09
F-values and LSD$_t$	0.05:	1986		1987
	Wheat	172***, 0.05		38.13***, 0.06
	Barley	239***, 0.05		2.9*, 0.11

end of the experiment — 24 weeks. The result was also the same for seed artificially infected with *B. sorokiniana,* but after 3 weeks the result was always inferior to that for uninfected seed (Fig. 1).

DISCUSSION

The powdery microbial preparation made from *S. griseovirides* bacteria was successful as a dusting agent on wheat and barley against foot rot damage caused by *Fusarium* spp. and *B. sorokiniana* fungus in pot experiments carried out in the greenhouse. However, the control result was significantly less than that obtained with chemical mercury dusting. This result is inferior to the results obtained in preliminary experiments in which *S. griseovirides* was found to be an effective antagonist against several pathogenic fungi on both nutrient media and in pot experiments. It also gave good control results against e.g. *F. culmorum* on cereals in pot experiments (TAHVONEN 1982). The methods employed in the earlier experiments are different from those used here, and hence direct comparison of the results is not possible.

Fusarium spp. and *B. sorokiniana* are rather common seed-borne moulds on cereals in Finland (UOTI and YLIMÄKI 1974, KURPPA 1984), and they also occurred abundantly in the seed lots used in these experiments. Compared to chemical control, these results are relatively less successful than the corresponding results obtained with cruciferous plants (TAHVONEN and AVIKAINEN 1987). A number of the moulds occurring on cereals, especially *B. sorokiniana,* are able to penetrate into the inner parts of the seed (KURPPA 1984). The antagonist acting on the surface of the seed thus does not come into complete contact with the pathogen. As the seeds of cruciferous plants are smooth, and seed-borne moulds are found only on the surface of the seed (NEEGAARD 1945, TAHVONEN 1979), it is obviously easier for the antagonist to function on these plants than on cereals.

The control effect obtained with a dusting dose of 5 g/kg was almost as good as that given by 10—15 g/kg which, in practice, were difficult to apply. The retention of effectiveness is in agreement with the results of experiments carried out earlier with cruciferous seed. Similarly, the effectiveness of the dusting effect on stored seed lasted for a similar period to that

obtained in earlier experiments (TAHVONEN and AVIKAINEN 1987). The partial retention of the dusting effectiveness for a number of weeks and months demonstrated the viability of the powdery *Streptomyces* preparation. The antagonist was found to grow on all the dusted seed, from all the sown replications, when the seeds were germinated on filter paper. The presence of some of the active compound produced by the antagonist in the preparation may also have reduced the extent of the disease.

The negative effect of *Streptomyces* bacteria on the germination of barley seed on moist filter paper was a new observation which has not been reported in studies carried out earlier on cruciferous plants (TAHVONEN 1982, TAHVONEN and AVIKAINEN 1987). However, the dusted seeds sprouted in peat to the same extent as the control seed and those dusted chemically. No corresponding inhibition of germination was, however, found for wheat. The reason for the difference between wheat and barley in the germination of seed dusted with *Streptomyces* is unknown. Similar inhibition of germination has earlier been reported for lettuce, carrot, dill and parsley, but shoot formation *in vivo* has always been normal (TAHVONEN and AVIKAINEN, unpubl. data).

Trichoderma and *Gliocladium* species have been most studied in the biological control of root pathogens of plants, especially in the control of *Fusarium* and *Gaeumannomyces* fungi

(DOMS and GAMS 1968, DENNIS and WEBSTER 1971 a, b). *Pseudomonas* bacteria have attracted special attention in the control of take-all on wheat (WELLER 1983, WELLER and COOK 1983). The microbes used in the biological control of these cereal pathogens have, without exception, been isolated from agricultural soil. In the study presented here, however, only isolates from light, partly decomposed peat were used. This feature makes the field experiments interesting because there is little information about how *Streptomyces* adapts to the roots of cereals compared to its normal environment.

So far, relatively little work has been done on the use of *Streptomyces* bacteria in biological control on cereals. MERRIMAN et al. (1974 a, b) have studied the control of *Rhizoctonia solani* fungus on cereals. The main emphasis in other studies has been on laboratory experiments and ecological investigations (DOMSCH and GAMS 1968, TURHAN 1981 a, b).

According to the pot experiments, *Streptomyces* also has potential use as a biological control agent against cereal pathogens, although under field conditions the control result is hardly likely to exceed that achieved with chemical control, and its effectiveness is not sufficient against those pathogens which penetrate into the seed. The results of field experiments are being treated in another part of the study, where the properties of and potential of *Streptomyces* dusting can be seen under field conditions.

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SELOSTUS

Streptomyces sp. -torjuntapieneliön vaikutus vehnän ja ohran siemenlevintäisiin tyvitauteihin

I. Astiakokeet

RISTO TAHVONEN ja HANNA AVIKAINEN

Maatalouden tutkimuskeskus

Streptomyces griseovirides -mikrobista valmistettua jauhemaista preparaattia, jonka kaupallisena nimenä on Mycostop, käytettiin 3—15 g/kg siemenen peittauksena vehnällä ja ohralla torjumaan siemenlevintäiset *Bipolaris sorokiniana* ja *Fusarium* spp. -sienet astiakokeissa. Verranteena biologiselle peittaukselle oli siementen käsittely alkoksialkyylielohopeavalmisteella. Kasvatukset tehtiin kasvihuoneissa vaaleassa kasvuturpeessa. Osa kokeista tehtiin keinosaastutetuilla siemenillä.

Keinosaastutettujen siementen peittaus *Streptomyces*-valmisteella vähensi erittäin merkitsevästi *F. culmorum* ja *B. sorokiniana* sienten aiheuttamaa viotusta ohralla, mutta kemiallinen peittaus antoi aina paremman tuloksen kuin biologinen peittaus. Sekä biologinen että kemiallinen peittaus lisäsi oraiden kuivapainoja.

Luontaisesti saastuneilla siemenillä peittaukset vähensivät orasvioituksia. Idätyskokeissa paperialustoilla *Streptomyces* -peittaus vähensi siemenistä *B. sorakiniana* ja *Fusarium* -pitoisuuksia. Ohran siementen itävyys laski, mutta vehnällä tätä ilmiötä ei todettu. Samat siemenet orastuivat kuitenkin astiakokeissa kontrollin tai elohopeapeittauksen veroisesti. *Streptomyces* -peittauksen teho säilyi peitatuilla siemenillä huoneen lämmössä varastoituna 3—4 viikkoa, minkä jälkeeṛ peittauksen teho aleni.

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CHEMICAL CONTROL OF EUROPEAN RED SPIDER MITE
PANONYCHUS ULMI (KOCH)

II. EVALUATION OF CLOFENTEZINE AND HEXYTHIAZOX

TUOMO TUOVINEN

TUOVINEN, T. 1990. Chemical control of European red spider mite *Panonychus ulmi* (Koch). II. Evaluation of clofentezine and hexythiazox. Ann. Agric. Fenn. 29: 195—204. (Agric. Res. Centre, Dept. Plant Protect., SF-31600 Jokioinen, Finland.)

In laboratory tests, 250 and 500 ppm clofentezine sprayed on winter eggs of *P. ulmi* at 0—63 day-degrees (dd) above + 7 °C, had a 68—92 % effect. If sprayed just before the beginning of egg hatching (128 dd above 7 °C), the effect was only 35 %. In field tests, a good effect was obtained when clofentezine was sprayed before the beginning of embryonic development of winter eggs.

In laboratory tests, 50 and 100 ppm hexythiazox diminished hatching of undeveloped winter eggs (92 and 99 % effect), but the effect was poor when sprayed after some development of the eggs had occurred (77 dd above 7 °C). In field tests, hexythiazox had a good effect when sprayed in spring during the winter egg hatching period or in July.

When sprayed five times on trees with low density populations of *P. ulmi,* in coordination with the apple scab spraying schedule, clofentezine and hexythiazox significantly diminished the numbers of phytoseiid mites, but did not totally eliminate them. Single summer treatments with both acaricides were relatively harmless on phytoseiid mites *Euseius finlandicus* and *Phytoseius macropilis.* Repeated summer sprays with clofentezine reduced numbers of *Aculus sclechtendali,* but hexythiazox did not have any effect on eriophyiid mites.

Index words: chemical control, acaricides, clofentezine, hexythiazox, European red spider mite, *Panonychus ulmi,* Eriophyidae, Phytoseiidae, *Euseius finlandicus, Phytoseius macropilis.*

INTRODUCTION

The results of the control experiments on the European red spider mite (ERM), *Panonychus ulmi* (Koch) (Acari: Tetranychidae), using flubenzimine compared to the conventional acaricides chinomethionate and dicofol as well as oxydemetonmethyl, have been published earlier (TUOVINEN 1989). Of the other available acaricides, ovicidal oil preparates have been widely used against ERM. Most of the ovicidal acaricides have been tar oil or various petroleum oil formulations. Sprays on winter eggs, before onset of the vegetation period, result in satisfactory control provided the coverage of the spray is complete and the egg hatching period short (van de VRIE 1985). In Finland, the hatching of winter eggs lasts several weeks (LISTO et al. 1939) and this probably is the reason for the often poor effect of mineral oil

preparates. The use of tar oils which have a good effect on ERM winter eggs is now prohibited in Finland because of the harmful compounds in these oils.

Recently, two new ovo-larvicidal compounds completely different in chemical structure as well as in mode of action compared to earlier acaricides have been introduced: clofentezine, effective primarily against eggs (BRYAN et al. 1981, NEAL et al. 1986) and hexythiazox, effective against the eggs and larvae of tetranychid mites (WELTY et al. 1988). These two compounds have been tested in laboratory and field experiments in order to evaluate their effectiveness against ERM and their impact on other mite groups in apple trees. In this study, the results of the above tests are reported and the use of clofentezine and hexythiazox compared to other acaricides is discussed.

MATERIAL AND METHODS

Clofentezine was used as 50 % WP formulation Apollo, produced by Schering AG, and hexythiazox as 10 % WP formulation Nissorun 10 WP, produced by Nippon Soda Co.

As reference products, a mineral oil formulation (Ovipron, BP), chinomethionate (25 % WP formulation Morestan, Bayer AG), flubenzimine (50 % WP formulation Cropotex, Bayer AG) and oxydemetonmethyl (26.5 % EC formulation Metasystox, Bayer AG) were used. In some of the field experiments, insecticides and fungicides were applied following normal spraying schedules. These sprays were carried out using the recommended concentrations and doses.

Laboratory experiments

Twigs containing ERM winter eggs were sampled from orchards during winter and were stored in 0— +3 °C before tests. For each test, twigs from the same orchard were used. 5—10 pieces 1—3 cm in length, halved twig bits containing 25—50 eggs each were put into petri dishes on filter paper and sprayed with 2 ml of water diluted preparate in a Potter tower. Control dishes were sprayed with pure water. After spraying, the twig bits were put on petri dishes and each bit circled by insect glue. The dishes were preserved in a growing chamber at +20/ +15 °C temperature, 75 ± 10 % Rh and 13/11 h photoperiod (L/D). The dishes were uncovered. Each treatment was replicated four times and control dishes were included. The twig bits were checked two and four weeks after the treatments and the hatched larvae stuck in the insect glue were counted.

Clofentezine at 0.025 and 0.05 % a.i. dilutions was tested using eggs at various developmental stages. Eggs were obtained by preserving twigs in 0 °C, +5 °C, +10 °C and +15 °C for 0—21 days. Hexythiazox was tested on undeveloped and partly developed eggs using 0.005 and 0.01 % a.i. dilutions.

During laboratory experiments in growing chambers, the temperature sums were recorded using a growing degree day accumulator (TA51-P, Omnidata Int. Inc.). As a threshold temperature for winter egg development, +5 and +7 °C was used (LEES 1953).

Field experiments

In a commercial orchard, Paimio 1986—88, clofentezine was compared with chinomethionate and flubenzimine in two 0.5 ha blocks. In Piikkiö, 1988, hexythiazox was compared with chinomethionate in a demonstrative. test. In both orchards, insecticides against moths and

fungicides against the apple scab *Venturia inaequalis* (Cooke) Winter were also used.

In an experimental orchard, Jokioinen 1988, hexythiazox and clofentezine together with flubenzimine, were tested using fully randomized design and six single tree replicates. Preparates were sprayed according to the apple scab spraying schedule using lower concentrations in order to check the possible effect of these acaricides on apple scab. As a reference product bitertanol (Baykor, Bayer AG) was sprayed against the apple scab.

In an experimental orchard, Pälkäne 1988, summer applications of hexythiazox were studied using randomized block design and three single tree replicates. In 1989, the effect of early sprays of clofentezine, hexythiazox and a mineral oil preparate was studied.

In Paimio and Piikkiö the effect of sprays was checked 1—4 times during summer by sampling 5 leaves of equal size and position from 10—20 randomly selected trees, and in the au-

tumn, by sampling five 20 cm twig pieces from 10—20 trees. In Jokioinen, 20 leaves from each tree were sampled in July, and in Pälkäne, 10 leaf-rosettes in spring and 10 leaves during summer from each tree were sampled. Numbers of living mobile mites and ERM eggs were counted under a stereomicroscope. All relevant mite groups, including Tetranychidae, Tydeidae, Phytoseiidae and Eriophyidae, were observed. In some cases, numbers of eriophyids were estimated using a scale from 0 (= no mites) to 3 (= over 100 mites/leaf).

Meteorological data were obtained from the nearest meteorological station (Piikkiö and Pälkäne). Cumulative temperature sums over + 5 °C (day-degrees) were calculated for timing of the sprays. Data from laboratory tests were analysed using the analysis of variance (Tukey's test) and from the field experiments using either t-test or analysis of variance (Duncan's multiple range test) on $\log(x + 1)$-transformed data (STEEL and TORRIE 1980).

<center>RESULTS</center>

Laboratory experiments

The temperature sums needed for the beginning of ERM winter egg hatching were 140 day-degrees for + 7 °C and 170 dd for + 5 °C threshold temperatures (Fig. 1). Half of the eggs were hatched when 170 and 200 dd above + 7 and + 5 °C were reached, respectively.
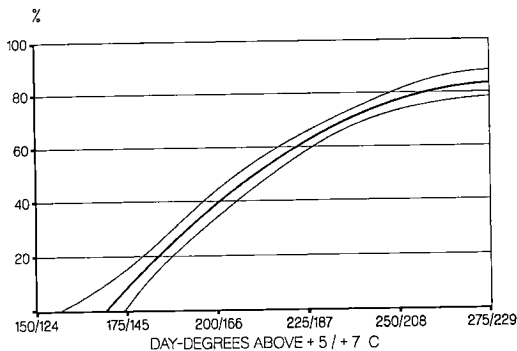

Fig. 1. Hatching of ERM winter eggs in a growing chamber in + 20/15 °C, 13/11 h photoperiod (L/D) and 75 ± 10 % Rh. Day-degrees recorded from the beginning of the test. Regression curve calculated from 19 replicates, 95 % confidence intervals are included.

Table 1. Effect of clofentezine on ERM winter eggs at different stages of egg development. Treatments in Potter tower, 2 ml of dilution/replicate (see text).

Preserving		dd (7 °C)	Effect % (Abbott)	
temp. (°C)	time (d)		250 ppm	500 ppm
0	7	0	87.3[b]	91.5[b]
5	14	0	92.2[b]	90.7[b]
5	28	0	68.4[b]	81.0[b]
10	7	21	77.9[b]	82.0[b]
10	14	42	83.4[b]	92.5[b]
10	21	63	77.2[b]	79.8[b]
15	16	128	37.3[a]	35.1[a]

Means with different letters in columns denote significant differences (P < 0.05) according to Tukey's test.

Clofentezine affected undeveloped ERM winter eggs well (Table 1). As embryonic development progressed the effect of clofentezine on egg hatching diminished when sprayed 1—2 days before hatching. The concentration of 250 ppm was only slightly less effective than the double one.

Hexythiazox at 50 and 100 ppm concentration killed the undeveloped winter eggs almost totally but, like clofentezine, the effect on the more developed eggs was poor (Table 2).

Table 2. Effect (Abbott) of hexythiazox and clofentezine on ERM winter eggs. Exp. 1. was carried out on undeveloped eggs, Exp. 2. on winter eggs after preserving in +20/15 °C for 7 days (=77 dd above 7 °C). Treatments as in Table 1.

Treatment (% a.i.)	Effect % (hatch. %)			
	Exp. 1.		Exp. 2.	
Hexythiazox (0.005)	99.3	(0.6ª)	24.4	(70.5ᵇ)
Hexythiazox (0.01)	99.4	(0.5ª)	48.9	(47.6ª)
Clofentezine (0.025)	92.6	(6.2ᵇ)	34.9	(60.7ᵇ)
Untreated		(84.1ᶜ)		(93.3ᶜ)

Means with different letters in columns indicate significant differences (P < 0.05) in hatching-% according to Tukey's test.

Field experiments

In 1986, Paimio, clofentezine was sprayed on winter eggs, a few of which had already hatched (9.5., sprayed when 83 day-degrees above +5 °C was reached). The initial ERM winter egg density was high, 199 eggs/10 cm branch, leading to a high population level on the control block. At first, in June, the effect of clofentezine was satisfactory compared to the control block, but in July and August, an outbreak of ERM occurred (Table 3). A spray

Table 3. Results of the field experiment in Paimio, 1986. Blocks of about 0.5 ha were sprayed with a mist sprayer, 300 l/ha. Numbers of mobile mites and eggs were counted from 5 leaves/tree, and numbers of winter eggs from 5 twigs/tree from 10 trees/treatment. Before the treatment in spring the number of winter eggs was 199/10 cm twig.

Treatment and rate (g a.i./100 l)	Date	No. of ERM/leaf (mobile and eggs)						ERM winter-eggs
		29.5. mob	19.6 mob	23.7		16.9		
				mob	eggs	mob	eggs	
Clofentezine (100)	9.5.	8.3	1.1	29.4	43.2	75.1	34.8	219
Chinomethionate (62.5)	4.6.	104.5	1.9	70.5	75.8	68.3	46.0	267
T-test		***	NS	***	***	NS	*	NS

Other treatments in the area: fenitrothion (375 g a.i./100 l) 20.5. (only clofentezine) and 19.6., dimethoate (133) 7.7., dithianon (225) 14.5., 23.5., 4.6., 9.6., 19.6., 13.7. and triforine (123.5) 7.7. (for scab control). * P < 0.05; *** P < 0.001

Table 4. Results of the field experiment in Paimio, 1987. Blocks of about 0.5 ha were sprayed with a mist sprayer, 300 l/ha. Numbers of ERM were counted from 5 leaves/tree, and numbers of winter eggs from 5 twigs/tree from 10 trees/treatment. Before the sprays in spring the number of winter eggs was 243/10 cm twig.

Treatment and rate (g a.i./100 l)	Date	No. of ERM/leaf (mobile and eggs)						ERM winter-eggs
		22.6. mob	16.7.		30.7.		Erioph.	
			mob	eggs	mob	eggs		
Clofentezine (100)	29.4.	0.04	0.22	0.32	0.31	1.3	>100	—¹
Flubenzimine (250)	10.6.	—	0.12	0.33	1.6	3.9	0	17.5
T-test			NS	NS	*	NS	—	

¹ — sign indicates missing data.

Other treatments in the whole area: dimethoate (133 g a.i./100 l) 3.6. and 22.7., dithianon (250) 18.5., 3.6., 10.6., 14.6., 23.6., 29.6. and 9.7. (for scab control). * P < 0.05

Table 5. Results of the field experiment in Paimio, 1988. Blocks of about 0.5 ha were sprayed with a tractor mist sprayer, 400 l/ha. Numbers of ERM were counted from 100 leaves/treatment and winter eggs from 20 twigs/treatment.

Treatment and rate (g a.i./100 l)	Date	No. of ERM/leaf (mobile and eggs)						
		15.6.		30.6.		Erioph.	10.8.	
		mob	eggs	mob	eggs		mob	eggs
Hexythiazox (15.0)+ chinomethionate (62.5)	19.5. + 4.7., 14.7.	0.04	0.4	1.0	2.2	71.1	0.0	0.0
Chinomethionate (62.5)	20.5., 4.7., 14.7.	0.03	7.2	13.7	38.1	40.6	0.0	0.3
T-test		NS	***	***	***	**	—	NS

Other treatments in whole area: dimethoate (133 g a.i./100 l) 20.—23.5., 4.7. and 14.7., dithianon (250) 23.5., 2.6., 9.6. and 21.6., bitertanol (100) 27.6. (for scab control). ** P<0.01, *** P<0.001

Table 6. Results of the field experiment in Piikkiö, 1988. Blocks of 0.25—0.5 ha were sprayed with a mist sprayer, 400 l/ha. Numbers of ERM were counted from 5 leaves/tree, and winter eggs from 10 twigs/treatment.

Treatment and rate (g a.i./100 l)	Date	No. of ERM/leaf				ERM winter-eggs
		8.6.		10.8.		
		mob	eggs	mob	eggs	
Hexythiazox (12.5)	24.5.	0.0	0.2	3.6	11.7	132
Hexythiazox (12.5)+ chinomethionate (62.5)	24.5., 3.6.	0.3	0.3	1.3	4.0	95
Chinomethionate (62.5)	27.5., 3.6.	0.2	0.6	4.0	23.1	—[1]

[1] not counted

Other treatments in whole area: fenitrothion (375 g a.i./100 l) 18.5., dimethoate (160) 30.6., bitertanol (125) 24.5., 3.6., 23.6., 30.6. (for scab control).

with chinomethionate before any summer eggs were laid (4.6., 225 dd) was also satisfactory, but later the numbers of ERM burgeoned. June and July were warmer than usual, favouring the outbreak of ERM. Only a few specimens of phytoseiid mites were found in this experiment and predatory insects were extremely scarce, too. Tydeids were not found and eriophyids were present only in low numbers.

In 1987, the effect of clofentezine was compared to that of flubenzimine in Paimio. Now, clofentezine was sprayed earlier (29.4., 0 dd) than in 1986 and winter eggs had not yet begun to develop. Both clofentezine and flubenzimine (10.6., 124 dd) had a good effect on ERM (Table 4). Clofentezine did not have any effect on the apple rust mite *Aculus schlechtendali*

(Nal.) (Acari: Eriophyidae), whereas no gall mites were found in the trees sprayed with flubenzimine. A few larvae of a dipterous predator *Arthrocnodax mali* Kieffer (Diptera: Cecidomyiidae) were found on leaves treated with clofentezine, but none on flubenzimine treated leaves. Exceptionally cold and rainy weather in summer 1987 (1030 dd during the whole season) suppressed ERM reproduction. However, in a neighbouring block, not included in the experiment, which was sprayed with chinomethionate (26.6.) mobile ERM numbers exceeded 10/leaf at the end of July.

In Paimio, 1988, the effect of one spray with hexythiazox was compared to chinometionate. A spring spray before the beginning of the egg hatching period (19.5., 89 dd), at first resulted

in good control but because of the very warm weather, the number of ERM grew quite high in the chinometionate block (Table 5). The two late treatments with chinometionate prevented a mite outbreak in July. Repeated treatments with chinometionate had some effect on eriophyids. Phytoseiid mites were not found in this experiment.

In Piikkiö, 1988, sprays with hexythiazox (24.5., 115 dd) and chinomethionate (27.5., 138 dd and 3.6., 200 dd) resulted in satisfactory control of ERM (Table 6). Eriophyids, tydeids or phytoseiids did not occur in experimental blocks. Summer 1988 was very warm (1564 dd during the whole season) which led to quite a high overwintering population of ERM.

In 1988, Jokioinen, clofentezine, hexythiazox and flubenzimine were sprayed in lower concentrations according to the apple scab spraying schedule (Table 7). The initial density of ERM was low, and the effect of sprays could not be confirmed, although the acaricidal treat-

Table 7. Results of the field experiment in Jokioinen, 1988. 6 apple trees/treatment were sprayed according to the apple scab spraying program, with a knapsack compression sprayer, on 23.5., 3.6. and 15.6., and with a knapsack mist sprayer on 21.6. and 27.6. Samples of 20 leaves/tree were checked 18.7. and number of mites were counted or estimated (Eriophyidae).

Treatment and rate (g a.i./100 l)	Number of mites/10 leaf			
	ERM		Erioph.	Phytos.
	mob	eggs		
Flubenzimine (3 × 25, 2 × 85)	0.0	0.0	3.0a	0.03a
Clofentezene (3 × 25, 2 × 85)	0.67	0.58	3.0a	3.5a
Hexythiazox (3 × 5, 2 × 17)	0.17	0.33	83.0b	2.33a
Bitertanol[1] (3 × 12.5, 2 × 42.5)	1.25	0.75	95.0b	19.5b
No treatments	1.17	0.75	67.0b	17.8b

[1] Fungicide used against the apple scab.

Means with different letters in columns denote significant differences (P < 0.05) according to Duncan's multiple range test on log-transformed data. Columns without letters indicate a nonsignificant F-test.

Table 8. Effect of summer sprays with hexythiazox on ERM population densities during the following season. A field experiment in Pälkäne, 1988 (Häme Res. Station). Sprays were performed using a knapsack mist sprayer, 0.5 l/tree. Numbers of mites were counted from 5 leaf-rosettes (19.5.) or from 10 leaves/tree and ERM winter eggs from 5 twigs/tree. No. of Eriophyids were estimated using a scale from 0 to 3 (see text). Tyd = Tydeidae, Eriop = Eriophyidae, Phyt = Phytoseiidae.

Treatment and rate (g a.i./100 l)	Date	88/89 Winter eggs	19.5.89 (ros.)			8.6.89					18.7.89				
			ERM mob	Tyd	Phyt	ERM mob	eggs	Tyd	Eriop	Phyt	ERM mob	eggs	Tyd	Eriop	Phyt
Hexythiazox (15.0)	6.7.	3.8b	0.0a	0.9a	0.1	0.0a	0.2a	0.9b	1.1	0.2ab	0.1a	2.3b	2.5b	0.4	2.3b
Hexythiazox (15.0)	20.7.	0.9a	0.0a	0.0a	0.0	0.0a	0.1a	0.1a	1.0	0.3bc	0.3a	2.3ab	1.2a	0.5	2.3b
Hexythiazox (15.0)	6.7.+ 20.7.	0.1a	0.0a	0.0a	0.0	0.0a	0.0a	0.2a	0.7	0.7c	0.2a	0.2a	1.3a	0.1	4.4b
No treatment		215.3c	147.7b	11.7b	0.0	11.4b	169.9b	7.0c	0.2a	0.0a	8.4b	22.1c	10.2c	0.4	0.4a

Means with different letters in columns denote significant differences (P < 0.05) according to Duncan's multiple range test on log-transformed data. Columns without letters indicate a nonsignificant F-test. No test was calculated for Eriophyidae.

Table 9. Field experiment in Pälkäne, 1989 (Häme Res. Station). Sprays were performed on 25.4. with a knapsack mist sprayer, 0.5 l/tree. For counting of mites, see Table 8.

Treatment and rate (g a.i./100 l)	88/89 Winter eggs	No. of mites/leaf (ERM: mobile and eggs)												
		19.5.89 (ros.)			8.6.89					18.7.89				
		ERM mob	Tyd	Phyt	ERM		Tyd	Eriop	Phyt	ERM		Tyd	Eriop	Phyt
					mob	eggs				mob	eggs			
Hexythiazox (10.0)	253.5	238.9^b	28.9^b	0.1	12.9^b	164.5^c	10.5^b	0.2	0.5^b	7.1	25.2	11.8^bc	0.7	0.8^b
Clofentezine (50.0)	256.9	11.5^a	38.4^b	0.1	0.5^a	11.4^a	15.5^b	0.2	0.0^a	3.9	6.2	8.3^ab	0.4	0.3^ab
Mineral oil (291)	148.3	56.4^b	11.5^a	0.0	1.2^a	32.3^b	1.8^a	0.2	0.0^a	5.1	18.2	5.9^a	1.2	0.1^a
No treatment	215.3	147.7^c	11.7^a	0.0	11.4^b	169.9^c	7.0^b	0.2	0.0^a	8.4	22.1	10.2^c	0.4	0.4^ab

Means with different letters in columns denote significant differences (P<0.05) according to Duncan's multiple range test on log-transformed data. Columns without letters indicate a nonsignificant F-test. No test was calculated for Eriophyidae.

ments reduced numbers of ERM. Clofentezine had a good effect on the apple rust mite but it did not affect the other mites present on the leaves, the most frequent species belonging to Tydeidae. Hexythiazox had no effect on eriohyids. Both clofentezine and hexythiazox diminished the numbers of phytoseiid mites *Euseius finlandicus* (Oud.) and *Phytoseius macropilis* (Banks) but not as dramatically as flubenzimine. Neither of the acaricides affected the apple scab. The fungicide bitertanol had no effect on mites.

In 1988, Pälkäne, one or two summer sprays with hexythiazox resulted in very low densities of overwintering ERM eggs (Table 8). One spray was as effective as two sprays with a two-week interval and timing of the sprays had only a slight effect, the later treatment being slightly better. Because of the very warm summer, ERM density in untreated trees grew quite high. Mite population densities were checked during the next summer and on the treated trees ERM population stayed very low although the weather conditions were favourable for mites. The numbers of phytoseiids and eriophyids were not lower than in control trees, but tydeid mites were less numerous. No other pesticides were used in this part of the orchard during 1988—89, which made it possible for predatory bugs, especially *Anthocoris nemorum* (L.) (Heteroptera: Anthocoridae), to be present in high numbers and feed on mites. The effect of anthocorids was clearly seen in control trees, where ERM numbers diminished although phytoseiids were quite scarce.

In 1989, early spring treatments (25.4., 0 dd) with clofentezine or mineral oil on a high population of overwintering ERM eggs resulted in good control (Table 9). Hexythiazox did not have any effect on ERM. None of the treatments lowered the number of phytoseiid mites; mineral oil had an adverse effect on tydeids.

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Results of the laboratory tests suggested the use of clofentezine early in spring, before or in the beginning of the embryonic development of eggs. Field tests confirmed this finding. BAIL-LOD et al. (1986), using 0.04 % a.i. clofentezine in laboratory tests on eggs 8 days before hatching, achieved better results at constant $+12\ °C$ temperature (97 % effect) than at $+15\ °C$ (74 %) or at $+20\ °C$ (32 %). These results also support a rather early use of clofentezine. However, BRYAN et al. (1981) recommend spray with clofentezine just before winter egg hatching, partly because of the residual effect of clofentezine on young larvae — nymphal and adult stages of ERM are not affected by clofentezine. The laboratory test method used herein did not reveal the residual effect on newly hatched larvae. With respect to the results of the field tests, the residual effect of clofentezine was clearly seen. Early sprays of clofentezine should be preferred when conditions are favourable, otherwise the spray at the beginning of egg hatching will result in good control, too. According to ROCK (1987), also a single summer application using 142—284 g a.i./ha led to good and long-lasting control of ERM.

The doses of clofentezine used in this study varied from 150 to 300 g a.i./ha. PEREGRINE et al. (1986) studied the influence of application volume in early spring sprays using a constant amount of 200 g a.i./ha. They concluded that clofentezine was effective on all tested volumes ranging from 100 to 2000 l/ha, but higher volumes resulted in more uniform results. In Finland, growers usually spray 200—400 l/ha, which seems to be sufficient when spring sprays are concerned.

In laboratory tests, hexythiazox was effective on undeveloped ERM winter eggs both with 50 and 100 ppm concentrations, but more developed eggs (77 dd above $+7\ °C$) were affected much less. In laboratory tests carried out by WELTY et al. (1988) they found that the effect of 100 ppm hexythiazox on eggs sprayed less than 2 days before hatching was only 52 %, compared with 90—93 % on less developed eggs. However, the residual effect of 100 ppm hexythiazox on newly hatched larvae was 94—100 % (WELTY et al. 1988).

In field tests, the effect of hexythiazox sprayed at the beginning of the egg hatching period was satisfactory, but an early spray before the beginning of egg development did not have any effect at all. The reason for this cannot be explained for certain, but e.g. weather conditions in the field after the treatment often reduce the effectiveness of pesticides compared to laboratory conditions. Rainfall may have influenced hexythiazox — in Pälkäne the 3rd and 4th days after the spray were rainy: 5.8 mm and 17.9 mm of rainfall, respectively. In conclusion, hexythiazox cannot be recommended for early spring sprays under Finnish conditions. The effect of clofentezine, sprayed at the same time, was very good, and also the mineral oil spray resulted in satisfactory control.

Hexythiazox (15 g a.i./100 l water, 300 l/ha) proved to be very good against ERM when sprayed once or twice in July. Also ROCK (1987) obtained a good effect with a single spray (71 g a.i./ha) in the beginning of June in North Carolina, USA. In the present study, during 1988—89, which was much warmer than normal, one spray in July was enough to maintain the population of ERM at quite a low level. However, this low level is certainly not only based on the effect of the acaricide, but also is a result of the activity of natural enemies. Phytoseiid mites were numerous in all the treated trees, and *Anthocoris* spp. bugs were also common. These tests demonstrated that omission of harmful pesticides allows natural enemies to maintain the population of ERM at a low level (c.f. KROPCZYNSKA and TUOVINEN 1988).

Clofentezine and hexythiazox, when sprayed five times during the season in lower concentrations, clearly diminished the number of phytoseiid mites. However, quite high populations of phytoseiids remained on the leaves, showing that the preparates were not very toxic to predatory mites. HOY and OUYANG (1986) noted that clofentezine and hexythiazox were less toxic to the eggs of a predatory mite *Metaseiulus occidentalis* Nesbitt than to the eggs of the tetranychid mites *Tetranychus pacificus* McGregor and *T. urticae* Koch. KARG et al. (1987) graded clofentezine as a harmless acaricide for a wide range of natural enemies. In their summary article on the side effects of pesticides, BOLLER et al. (1989) presented that both acaricides are harmless to *Typhlodromus pyri* Scheuten. In this study, the most common phytoseiid species were *Phytoseius macropilis* and *Paraseiulus soleiger*. Repeated sprays of clofentezine and hexythiazox most likely affected, to some extent, the eggs and larvae but not the adult or nymphal stages and thus caused the reduction in phytoseiid numbers compared with control trees. Normally these acaricides are sprayed only once per season. The reference acaricide, flubenzimine, was harmful to all mite groups inhabiting apple trees (cf. TUOVINEN 1989).

Hexythiazox and clofentezine are thought to be a solution to the resistance problems which have arisen when the selective organotin acaricides cyhexatin and fenbutatinoxide have been employed without taking advantage of their selectivity (CROFT et al. 1987). In Finland, organotin acaricides have not been used against ERM, but the future use of both clofentezine and hexythiazox should be even more circumspect than that of the conventional acaricides and unnecessary treatments should be avoided. This is necessary to prevent the occurrence of possible resistance problems for as long as possible. To achieve this goal, the monitoring of both ERM and phytoseiid populations should be included as an integral part of the commercial apple growing technique.

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SELOSTUS

Hedelmäpuupunkin kemiallinen torjunta

II. Clofentetsiini ja hexythiazox

TUOMO TUOVINEN

Maatalouden tutkimuskeskus

Hedelmäpuupunkin torjuntaan soveltuvista torjunta-aineista clofentetsiini ja hexythiazox poikkeavat vaikutustavaltaan meillä aikaisemmin käytössä olleista akarisideista. Molemmat valmisteet vaikuttavat ensi sijassa muna-asteisiin, sekä talvi- että kesämuniin, mutta myös vastakuoriutuneisiin toukkiin. Sen sijaan myöhempiin kehitysasteisiin vaikutus on heikko.

Laboratoriokokeissa todettiin clofentetsiinin ja hexythiazoxin tehoavan paremmin kehittymättömiin kuin pitkälle kehittyneisiin talvimuniin. Kenttäkokeissa clofentetsiinilla saatiin samansuuntaisia tuloksia: huhtikuun loppuun ajoitettu käsittely osoittautui tehokkaammaksi kuin juuri ennen kukinnan alkua toukokuussa tehty ruiskutus. Hexythiazoxin osalta tulokset kenttäkokeissa olivat osittain ristiriidassa laboratoriokokeiden antamien tulosten kanssa: myöhäinen kevätkäsittely johti parempaan tulokseen. Toisaalta hexythiazoxin käyttö keskikesällä osoittautui kaikkein tehokkaimmaksi ja vaikutti vielä seuraavan vuoden punk-

kirunsauteen. Clofentetsiinin sopiva tehoainemäärä omenapuilla on varhain keväällä 150—200 g/ha, hexythiazoxin 50 g/ha. Nestemääräksi sumuruiskulla riittää 300 l/ha, kesällä suositellaan suurempaa nestemäärää paremman kattavuuden saamiseksi.

Sekä clofentetsiini että hexythiazox osoittautuivat suhteellisen haitattomiksi omenapuilla esiintyville petopunkeille. Vaikutus oli vähäinen myös äkämäpunkkeihin. Kumpikaan valmiste ei ole haitallinen hyönteispedoille tai loisille eikä pölyttäville hyönteisille, joten ne soveltuvat hyvin integroidun torjunnan yhteydessä käytettäväksi. Molempien akarisidien käytön tulisi perustua havaintoihin hedelmäpuupunkin ja petopunkkien runsaudesta, jolloin käsittelyt voidaan suorittaa todellisen tarpeen mukaan. Näin saadaan parhaiten hyödynnettyä valmisteiden selektiiviset ominaisuudet ja luodaan edellytyksiä hedelmäpuupunkin luontaisten vihollisten toiminnalle.

EFFECT OF FOUR FUNGICIDES ON PHYTOPHAGOUS AND PREDATORY MITES ON APPLE TREES

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TUOVINEN, T. 1990. Effect of four fungicides on phytophagous and predatory mites on apple trees. Ann. Agric. Fenn. 29: 205—215. (Agric. Res. Centre, Inst. Pl. Protect., SF-31600 Jokioinen, Finland.)

In laboratory tests, 1000 ppm dichlofluanid and 240 ppm triforine sprayed on larvae of *Panonychus ulmi* either killed them or prevented their further development, but 125 ppm bitertanol and 450 ppm dithianon had only a slight effect on larvae. Triforine had a 75 % effect on *P. ulmi* winter eggs when sprayed a few days before hatching but no effect on undeveloped eggs. Bitertanol, dichlofluanid and dithianon had no influence on winter egg hatching.

In a field experiment, four sprays in June—July with bitertanol caused a slight increase in the numbers of *P. ulmi* but did not affect Eriophyidae, Tydeidae or Phytoseiidae. Sprays with dithianon were harmless to the phytoseiid mites *Paraseiulus soleiger* and *Amblyseius canadensis*. Dichlofluanid diminished numbers and prevented egg laying of *P. ulmi,* and proved to be harmful to Eriophyidae and Phytoseiidae. Triforine diminished the numbers of Eriophyidae and Phytoseiidae, but the effect was only temporary.

The apple scab fungicides bitertanol and dithianon have been judged to be safe in integrated control programs. The use of triforine is advisable in early sprays, whereas dichlofluanid treatments should be avoided, if phytoseiid mites are present.

Index words: *Panonychus ulmi,* Phytoseiidae, *Amblyseius canadensis, Amblyseius reductus, Paraseiulus soleiger,* Eriophyidae, Tydeidae, bitertanol, dichlofluanid, dithianon, triforine, integrated control, side effect of fungicides.

INTRODUCTION

In Finland, the apple scab *Venturia inaequalis* (Cooke) Winter is the only commercially important disease of apple. It is currently controlled by repeated sprays of bitertanol, dichlofluanid, dithianon or triforine. In commercial orchards, fungicides are sprayed according to the length of rainy weather and prevailing temperature using the table presented by MILLS and LAPLANTE (1951), typically 4—8 times in May-July. Dithianon has been the most commonly employed fungicide for years, but recently, bitertanol and triforine have partly replaced it. Dichlofluanid has been applied in smaller amounts.

Although the destructive effects of fungicides on pest species are desirable in pest control, their similar effects on useful arthropods cause conflicts in integrated pest management programs. Laboratory and field tests surveying the side effects of pesticides have been intensively conducted by the IOBC Working Group, "Pesticides and beneficial organisms" whose work has been recently summarized by BOLLER et al.

(1989). The results of these tests are useful when rating pesticides for integrated control purposes. For many pesticides, only data on the initial toxicity of the pestcides in the laboratory are available. In the case of apple scab fungicides, which might be sprayed more than 10 times a year, the results of such laboratory tests may not reveal all aspects of the side effects in the field.

Of the apple scab fungicides available in Finland, dichlofluanid and triforine have been shown to have certain side effects on tetranychid mites (KOLBE 1968, BABIKIR 1978) and on some predatory phytoseiid mites (KARG et al. 1973, STENSETH 1975). Some evidence of the possible harmful effect of bitertanol on the European red spider mite *Panonychus ulmi* (Koch) (Acari: Tetranychidae) has been presented (BIGGS and HAGLEY 1988), but so far, no comparative study on the side effects of the use of the above mentioned apple scab fungicides has been performed.

The aim of this study is to compare and discuss the importance of the possible side effects of the fungicides used in Finnish apple orchards on the mite fauna in apple trees. The European red spider mite (ERM), predatory mites (Acari: Phytoseiidae), the apple rust mite *Aculus schlechtendali* (Nalepa) (Acari: Eriophyidae) as well as tydeid mites (Acari: Tydeidae) have been considered.

MATERIAL AND METHODS

Bitertanol was used as a 25 % WP formulation (Baykor, DuPont), dichlofluanid as a 50 % WP formulation (Euparen, Bayer), dithianon as a 75 % WP formulation (Delan, Shell) and triforine as a 19 % WP formulation (Saprol, Shell). The reference product employed in laboratory tests against ERM, was an ovo-larvicidal acaricide hexythiazox as 10 % WP formulation (Nippon Soda Co.). All sprays were performed using the recommended concentrations and doses.

Effect on ERM winter eggs and larvae

Laboratory tests were carried out on ERM winter eggs and larvae. For the test on winter eggs, they were preserved before the treatments for 10 days in a growing chamber at +20/15 °C temperature, 75 ± 10 % Rh and 16/8 h photoperiod so that the first eggs would have hatched 2—3 days after the treatments. Five to ten small twig bits, containing 25—50 winter eggs each, were placed onto filter papers in petri dishes and sprayed in a Potter tower using 2 ml diluted fungicide per treatment. Each treatment was replicated 4 times. Control dishes sprayed with water were included. After the treatments, the twig bits were moved onto petri dishes without filter paper and each bit encircled by insect glue (Oecotak). The twig bits were checked two and four weeks after the treatments by counting the hatched larvae that stuck in the glue circle.

The tests on larvae were carried out under one-day-old first generation ERM larvae, which were put on apple leaves, 25/leaf, using an artist's brush. The leaves were placed on wet foam rubber, face side down tightly on the rubber to prevent the larvae from hiding under the leaf. After one hour, when the larvae had started to feed, the leaves on foam rubber were sprayed in a Potter tower, and then the foam rubber pieces were put into water filled dishes so that each rubber was surrounded by water thus preventing the mites from escaping. Each treatment was replicated four times, and control leaves were sprayed with pure water. After the treatments, the dishes containing the leaves were stored in a growing chamber

(+20/15 °C, 75±10 % Rh, 16/8 h photo-period). Leaves were checked at 1—4 day intervals and the development of the mites was followed during 11—13 days.

Field experiment

In 1989, a field experiment was carried out in an old, neglected 0.5 ha orchard in Tammela, 100 km north of Helsinki. For each fungicide, one row was selected, leaving shelter rows between the sprayed ones. A spring spray with paraffin oil (Sun 7 E) for control of the apple sucker *Psylla mali* (Schmiedb.) (Homoptera: Psyllidae) was performed in the whole orchard before the ERM's winter egg hatching. This treatment obviously reduced also ERM numbers to some extent.

The fungicides were sprayed four times, on June 8, 16, and 22, and July 10, using a knapsack mist sprayer, the water amount being 1—1.5 l/tree. The total amounts of active ingredients per ha in the four treatments were as follows: bitertanol 1.1 kg, dichlofluanid 11.25 kg, dithianon 4.1 kg and triforine 2.1 kg.

Assessment of the effect of sprays was made four days after the second and eleven days after the fourth spray, and in the beginning of September. For assessment, 10 leaves from five trees in each treatment were sampled and inspected under a stereomicroscope. On each leaf, mobile and egg stages of ERM, mobile phytoseiid and tydeid mites were counted, and numbers of eriophyid mites estimated. Phytoseiid mites were prepared and the species identified using the key of MIEDEMA (1987) and with the help of a reference collection provided by T. Edland and the collection of KROPCZYNSKA and TUOVINEN (1988). Later in the winter, five 2—3 year-old twigs per tree from 5 trees/treatment were sampled to count ERM winter eggs.

<div style="text-align:center">RESULTS</div>

Effect on ERM winter eggs and larvae

Of the fungicides tested, only triforine (240 ppm) had a significant effect on ERM winter eggs applied close to hatch (Table 1). The slight reduction in hatching caused by dichlofluanid (1 000 ppm) and bitertanol (125 ppm) was not significant compared to untreated eggs. When sprayed on undeveloped winter eggs, triforine did not prevent them from hatching.

Triforine killed ERM larvae almost completely (Fig. 1). The few surviving larvae did not develop to nymphal stages. Dichlofluanid prevented the development of larvae although the killing effect was lower (P<0.001, Tukey's test) than that of triforine. Bitertanol and dithianon had no significant harmful effect on larvae, but the sprays caused more drowning in the surrounding water than did sprays with pure water. Both treatments caused a slight delay in nymphal development. The loss of animals in the course of the experimental period was 15—25 %, with the exception of bitertanol

Table 1. Effect of fungicides on ERM winter eggs in laboratory tests. A: test was carried out using long developed eggs, B: test was carried out using undeveloped eggs.

Treatment	A.i. ppm	Hatching %	Effect % (Abbott)
A.			
Bitertanol	125	60.1[a]	0
Dichlofluanid	1000	50.1[a]	15.0
Dithianon	450	49.1[a]	16.6
Triforine	240	14.7[b]	75.1
Untreated		58.9[a]	
B.			
Triforine	240	85.5	0
Untreated		80.1	

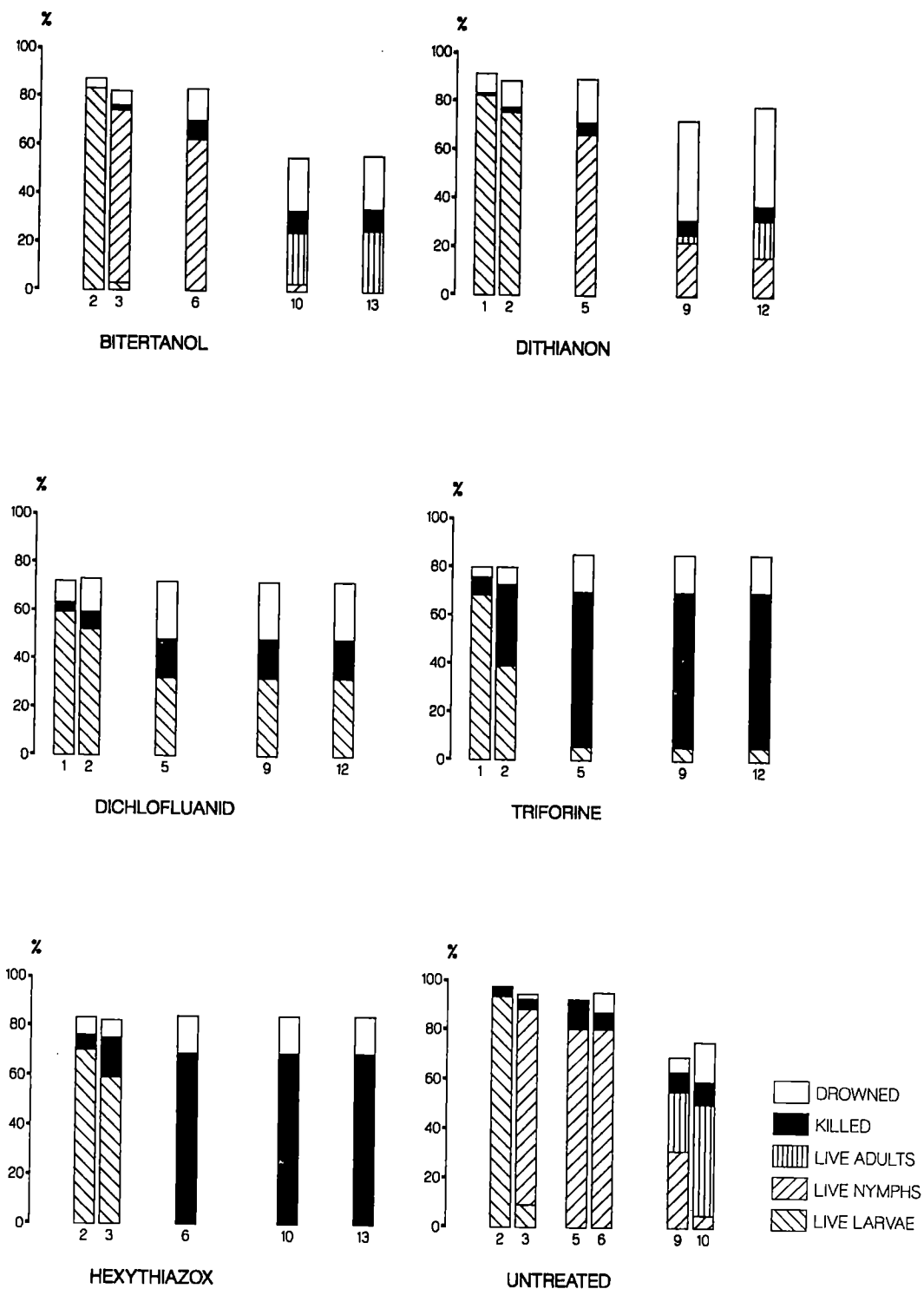Different letters denote significant differences (Tukey's test, P<0.05) in hatching % (A).

Fig. 1. Effect of fungicides on ERM larvae in the laboratory. Development of treated larvae during 10—13 days.
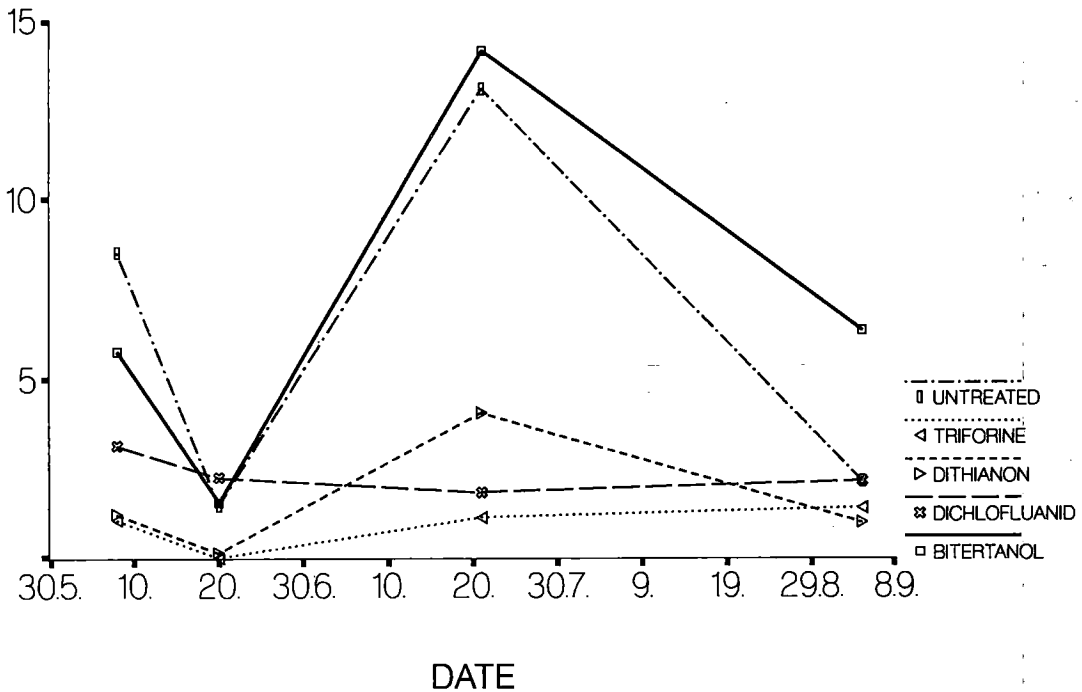
MOBILE P. ULMI / LEAF

Fig. 2. Effect of four sprays with fungicides (June 8.—July 10.) on numbers of mobile ERM.

treatments, where 43 % of mites disappeared from the test units.

Field experiment

Because of the differences in ERM numbers at the beginning of the experiment, the ERM data must be compared to the initial numbers before sprays to verify the population trends in each treatment (Figs. 2 and 3). Almost all of the mobile mites before the treatments were adults of the first generation (Fig. 2).

Because only four inspections were performed during the summer, the whole picture for the peak numbers in each treatment and generation cannot be presented. However, the inspection after all sprays on July 21 was timed, based on the temperature sum recordings and earlier experiences, to fit near the peak num-

ber of adult mites of the second generation (Fig. 2). During the period between June 20 and July 21, the ERM population in trees treated with dichlofluanid continued to decrease, and in the trees treated with triforine, the population growth was much less than in trees treated with dithianon, having the same initial population size. Summer egg inspections revealed that the greatest reduction in egg numbers occurred in trees treated with dichlofluanid, and also triforine sprays diminished egg laying (Fig. 3).

The last inspection in September revealed that mobile ERM occurred in quite low numbers, about 2 mites/leaf, but in bitertanol treated trees the numbers were about 3 times higher than in the untreated trees (Fig. 2). Because phytoseiids were present in equal numbers in both treatments (Fig. 4), the difference cannot
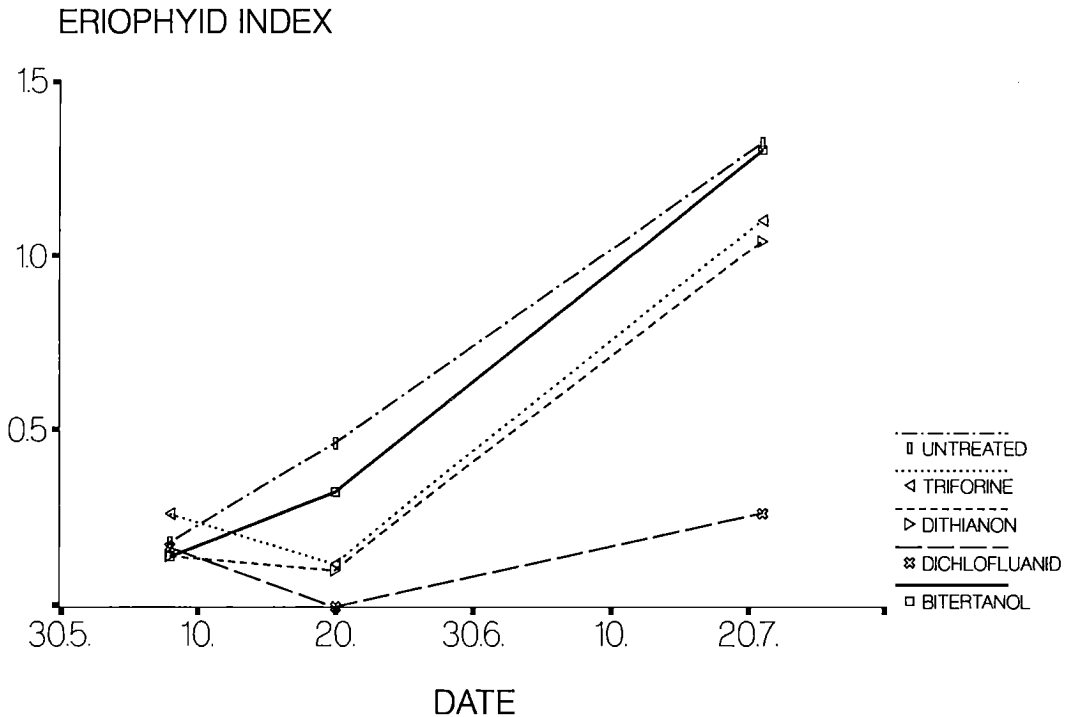
EGGS OF P. ULMI / LEAF
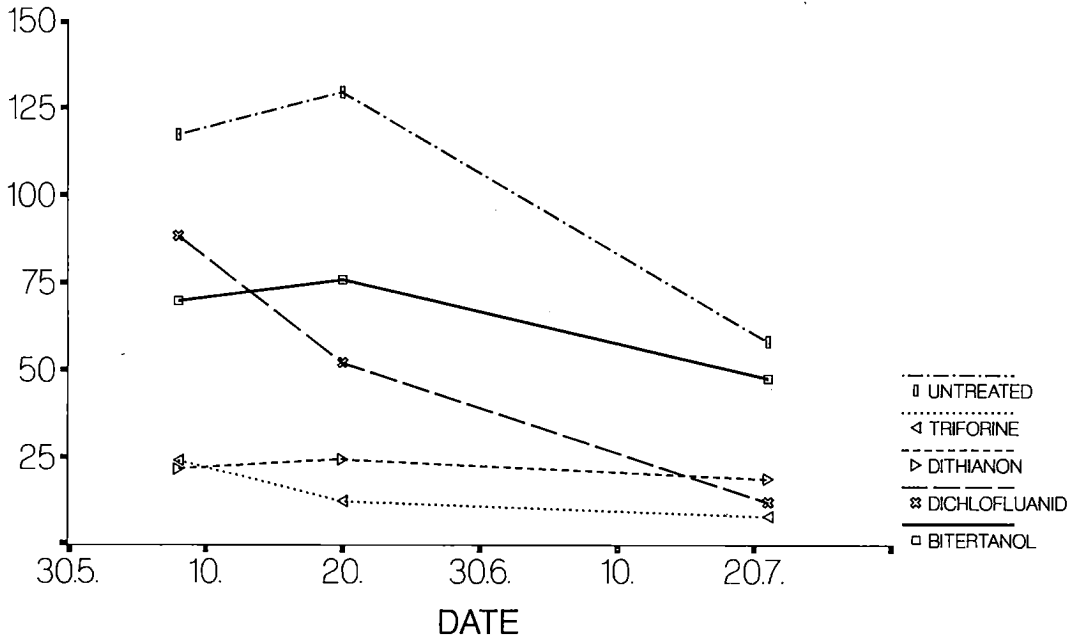
Fig. 3. Effect of four sprays with fungicides (June 8.—July 10.) on numbers of ERM eggs.

MOBILE PHYTOSEIIDS / LEAF
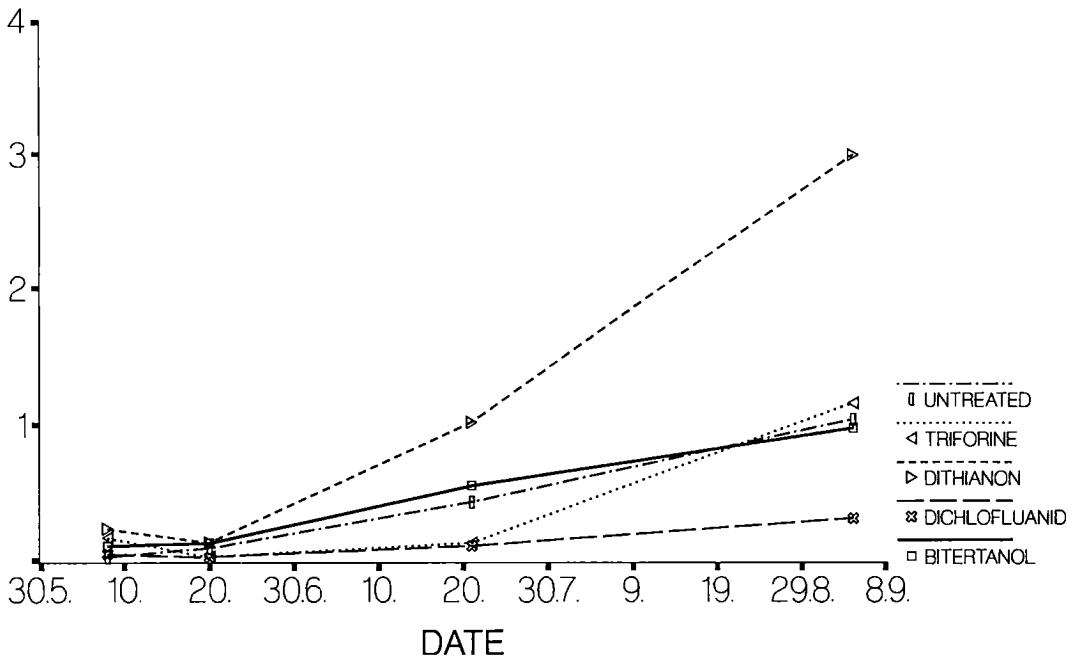
Fig. 4. Effect of four sprays with fungicides (June 8.—July 10.) on numbers of mobile phytoseiid mites.

Table 2. Effect of four fungicidal sprays in June—July on numbers of overwintering ERM eggs. Coefficient k describes the ERM population growth compared to that of untreated trees (= 1.0).

Treatment	Before treatment		k	After treatments:
	ERM/leaf			ERM winter eggs/10 cm
	adults	total		
Bitertanol	5.8	75.5	5.2	88.3
Dichlofluanid	3.1	91.2	0.8	7.3
Dithianon	1.2	23.1	3.8	13.3
Triforine	1.1	25.2	4.4	14.0
Untreated	8.5	125.7	1.0	25.0

be explained by the predation caused by phytoseiids in untreated trees.

Winter egg samples during the following winter showed that in trees treated with dichlofluanid, significantly less eggs were present than in untreated trees (P < 0.01, Student's t-test) or in trees treated with bitertanol (P < 0.05) (Ta-

ble 2). When the initial ERM population densities are taken into account, dichlofluanid caused a reduction in ERM numbers, especially when trees treated with other fungicides are compared. Looking at the adult mite numbers in September (Fig. 2), the low numbers of winter eggs in dichlofluanid treated trees suggest that a long-term effect, diminishing ERM fecundity may have been caused by dichlofluanid. The numbers of the apple sucker *P. mali* eggs were also counted, revealing that egg numbers in untreated trees were lower than in treated trees (P < 0.05).

The occurrence of mobile phytoseiids was scarce early in the season (eggs were not counted), increasing during the course of the summer, but present in significantly lower numbers in the trees sprayed with dichlofluanid compared to the control trees (P < 0.05), and the trees treated with dithianon (P < 0.001) or triforine (P < 0.05) (Fig. 4). The approximately

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Fig. 5. Effect of four sprays with fungicides (June 8.—July 10.) on numbers of eriophyid mites.

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MOBILE TYDEIDS / LEAF
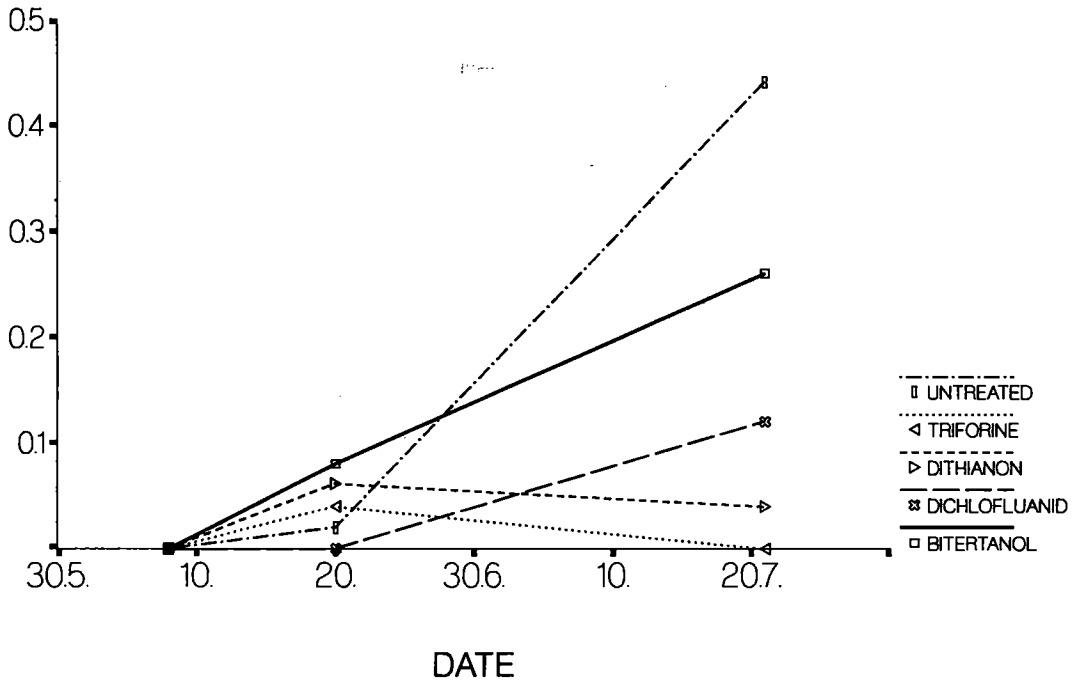
Fig. 6. Effect of four sprays with fungicides (June 8.—July 10.) on numbers of mobile tydeid mites.

three-fold number of phytoseiids in the trees treated with dithianon, compared to the other treatments (P < 0.01) may at least partly be explained by their greater numbers before the sprays. During the spraying period, dichlofluanid and triforine hindered phytoseiid population growth, but later the population rose even faster in triforine treated trees, whereas dichlofluanid caused a more permanent reduction in phytoseiid numbers.

Paraseiulus soleiger (Ribaga) was the most common of the altogether five species of phytoseiids found in the test samples (Table 3). The other four species were *Phytoseius macropilis* (Banks), *Euseius finlandicus* (Oud.), *Amblyseius reductus* (Wainstein) and *Amblyseius canadensis* (Chant and Hansell). The specimens identified as *A. cucumeris* (Oud.) in the study of KROPCZYNSKA and TUOVINEN (1988) appeared to be identical with *A. reductus* in the present study, therefore the earlier identification is considered incorrect.

Before the treatments, eriophyids occurred in almost equal numbers with the exception of

Table 3. The species composition (%) of phytoseiid mites in trees sprayed with various fungicides. Late summer inspection in 4.9.

Treatment	Total n	Ph. macropilis	E. finlandicus	A. reductus	A. canadensis	P. soleiger
Bitertanol	48	3	7	13	0	77
Dichlofluanid	16	0	0	20	0	80
Dithianon	156	0	0	1	23	76
Triforine	59	0	3	44	0	53
Untreated	52	8	0	0	17	75

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triforine block (Fig. 5). The first two sprays with dichlofluanid, dithianon and triforine caused a reduction in eriophyid numbers, but later, after four sprays, only in the trees treated with dichlofluanid were the numbers significantly lower ($P < 0.01$) than in other trees.

Tydeid mites, which were not identified to species, occurred only in low numbers in the experimental orchard (Fig. 6). The sprays, with the exception of bitertanol, seemed to have caused some reduction, although not significant, in the numbers of these mites.

DISCUSSION

Because of great differences in the initial numbers of ERM populations between treatments in the field experiment, only greater differences in population trends are considered to be meaningful. In case of phytoseiids as a group, the initial numbers in the experimental area were more homogenous. Comparisons between the species concerned are difficult, with the exception of *P. soleiger,* because of the diverse species spectrum in each treatment block.

Bitertanol is a relatively new fungicide (accepted for use in Finland in 1985), which has a long curative effect on apple scab, meaning that fewer treatments than with other available products may be possible. In the field experiment, bitertanol appeared to have no effect on ERM and other phytophagous mites, nor on phytoseiids, a fact which had been confirmed already in an earlier experiment (TUOVINEN 1989). Also BOLLER et al. (1989) have presented, based on laboratory and field tests, that bitertanol has no harmful effects on several beneficial organisms, including two phytoseiid species. BIGGS and HAGLEY (1988) concluded that 168 ppm bitertanol sprays, repeated at 14-day intervals, might suppress ERM population. Their results, however, do not support that conclusion, because the initial ERM numbers before the treatments were lower than in other treatments, except untreated trees, and later, the numbers did not differ from those of the untreated trees.

Dichlofluanid is commonly used against grey mold in strawberries and vegetables and less

against the apple scab. It has a clear effect on tetranychid mites (KOLBE 1968, KARG et al. 1973, SØRUM 1976) and this effect was shown also in the present laboratory tests. In the field experiment, dichlofluanid prevented the growth of ERM population and reduced winter egg-laying. The effect of dichlofluanid on other mite groups was harmful, and lasted the whole season in all mite groups. KARG et al. (1973) has proven that dichlofluanid sprays are harmful to the phytoseiid mite *Euseius finlandicus,* and BOLLER et al. (1989) presented that it is harmful to *Typhlodromus pyri* (Scheuten). In the present study, differences between phytoseiid species with respect to the effect of dichlofluanid could not be found. This fungicide is rated as harmless to anthocorid bugs (Heteroptera: Anthocoridae), which are important predators in apple orchards (BOLLER et al. 1989).

The most commonly employed fungicide against the apple scab in Finland, dithianon, appeared to have no effect on either ERM or other mites occurring on apple trees. Although laboratory tests showed that dithianon might have a sligt effect on ERM larvae, under field conditions the possible effect in question could not be verified. Dithianon showed a slightly harmful effect on eriophyid and tydeid mites. The data from the field experiment suggest that dithianon increased the numbers of the phytoseiids *P. soleiger* and *A. canadensis.* However, the nature of the mechanism of such a possible effect needs further study. Earlier studies

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have shown that the effect of dithianon is neutral when mites are concerned (FLEMMING et al. 1963, BOLLER et al. 1989).

Triforine has been reported to be effective against ERM (GILPATRICK et al. 1972), and slightly harmful to mobile stages of the phytoseiid mite *Phytoseiulus persimilis* Athias-Henriot (STENSETH 1975). However, van ZON and WYSOKI (1978) reported that 100—400 ppm triforine did not cause any mortality in females of the phytoseiid *P. persimilis*, nor in eggs or juveniles. In the present study, triforine was the only fungicide, which prevented ERM winter egg hatching, and in laboratory tests this fungicide was the most effective against ERM larvae. The results of the field test seem to support these results, although the low initial population level makes the comparison difficult. The population growth of phytoseiids was at first hindered by triforine, but after the sprays, the numbers of mobile phytoseiids grew faster compared to dichlofluanid. This observation suggests that triforine is not as harmful as dichlofluanid to all stages of phytoseiid mites, in this case *P. soleiger* and *A. reductus*. BOLLER et al. (1989) also presented that triforine is not harmful to *P. persimilis* or *T. pyri*. Eriophyid mites seem to recover from triforine treatments after the sprays have been discontinued.

In the field experiment, the overwintering ERM population in untreated trees appeared to be lower than could be expected on the basis of the size of the summer generation. As in this study no other predators than phytoseiid mites were collected, the effect of other factors could not be evaluated. At least anthocoriid bugs occurred as common predators in the orchard, but no comparative data are available to look at possible differences in their numbers between treatments.

CONCLUSIONS

Because of the observed differences in the fungicides used against apple scab concerning the side effects on ERM and phytoseiid mites, it is important to choose a fungicide also bearing in mind mite management in the apple orchard.

Only slight or no side effects were found in the case of bitertanol and dithianon, and these products are thought to be neutral as far as mite management is concerned.

Dichlofluanid should not be used for apple scab control, if the phytoseiids are to be conserved. On the other hand, dichlofluanid reduces ERM numbers so effectively that any special mite control might be unnecessary. Being harmless to predatory anthocorid bugs, this product might have restricted temporary use in integrated mite management, in situations when no phytoseiids are present.

Triforine affects all mite groups, but not as strongly as dichlofluanid. This fungicide, which also reduces ERM winter egg hatching, can be used in early spring sprays against apple scab. Phytoseiids, at least *P. soleiger* and *A. reductus*, seem to have some tolerance towards triforine, however, one should avoid frequent use of this fungicide.

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SELOSTUS

Omenaruven torjunta-aineiden vaikutus omenapuulla esiintyviin punkkeihin

TUOMO TUOVINEN

Maatalouden tutkimuskeskus

Suurin osa omenapuun kasvinsuojeluruiskutuksista on omenaruven torjuntaa. Suomessa tähän käytetään ditianoni-(Delan), bitertanoli- (Baykor), triforiini- (Saprol) ja diklofluanidi-valmisteita (Euparen). Toistuvilla ruiskutuksilla on vaikutuksia myös hyönteisiin ja punkkeihin. Tässä tutkimuksessa on selvitetty valmisteiden vaikutusta ensisijaisesti hedelmäpuupunkkiin ja petopunkkeihin.

Laboratoriokokeissa todettiin triforiinin heikentävän hedelmäpuupunkin talvimunien kuoriutumista, mikäli munat olivat saaneet kehittyä lähelle kuoriutumisajankohtaa. Muilla valmisteilla ei ollut vaikutusta talvimuniin. Triforiini ja diklofluanidi aiheuttivat hedelmäpuupunkin toukkien huomattavaa kuolleisuutta ja lisäksi estivät niiden yksilönkehitystä. Muiden valmisteiden vaikutus toukkiin oli vähäinen.

Kenttäkokeessa, jossa valmisteita ruiskutettiin neljä kertaa kesä-heinäkuussa, diklofluanidi häiritsi kaikkien punkkiryhmien lisääntymistä. Myös triforiini ehkäisi jossain määrin hedelmäpuupunkkien runsastumista, ja vaikutti aluksi haitallisesti myös petopunkkeihin ja äkämäpunkkeihin. Tämä vaikutus oli kuitenkin lyhytaikainen ja petopunkkikanta toipui nopeasti ruiskutuksista. Bitertanolilla ja ditianonilla ei ollut haitallista vaikutusta mihinkään punkkiryhmään, sen sijaan ditianonilla ruiskutetuissa puissa oli selvästi enemmän petopunkkeja kuin muissa puissa.

Käytännön johtopäätökset intgroidun torjunnan kannalta ovat seuraavat: 1. Bitertanolin ja ditianonin käyttö ei vaikuta haitallisesti punkkien luontaiseen torjuntaan petopunkkien avulla, mutta ei myöskään suoraan vähennä hedelmäpuupunkkien määrää. 2. Triforiinin käyttö keväällä ensimmäisissä ruiskutuksissa voi helpottaa hedelmäpuupunkin torjuntaa, aiheuttamatta kuitenkaan suurta vahinkoa petopunkeille. 3. Diklofluanidi vaikuttaa hedelmäpuupunkkia torjuvasti, mutta on haitallinen myös petopunkeille. Tilapäistä käyttöä voi harkita, jos petopunkkeja ei esiinny omenatarhassa.

FACTORS AFFECTING PANICLE PRODUCTION OF COKSFOOT (*DACTYLIS GLOMERATA* L.) IN FINLAND

I. DEVELOPMENT OF PANICLE PRODUCTION ABILITY AND TIME OF FLORAL INITIATION IN JOKIOINEN

OIVA NIEMELÄINEN

NIEMELÄINEN, O. 1990. Factors affecting panicle production of cocksfoot (*Dactylis glomerata* L.) in Finland. I. Development of panicle production ability and time of floral initiation in Jokioinen. Ann. Agric. Fenn. 29: 217—230. (Agric. Res. Centre, Inst. Crop and Soil Sci., SF-31600 Jokioinen, Finland.)

Development of the ability of cocksfoot cv. Haka to produce panicles was studied by transferring field grown sample plants into a glasshouse with conditions favourable to panicle growth during fall, winter and spring between September 1987 and May 1989 in Jokioinen (latitude 60°49′N), Finland.

The number of panicles produced per sample (a 15 cm long section of a row) was lowest in September (0—1.5), and highest in April (12.2—13.3) and in May (18.2.—22.0). Plants taken before December produced only a small number of panicles. Nevertheless panicle formation ability was present to some extent already in the fall but strengthened further during winter and spring. Panicle production ability in the fall was so small that the conditions during winter and spring have to be beneficial for panicle production to ensure sufficient panicle production for commercial seed production.

The critical daylength for floral initiation and for culm elongation was studied by growing plants at A) 10 h, B) 12 h, C) 14 h, and D) 16 h daylength in growth chambers for 9 weeks at +15 °C temperature. No panicles emerged in A and B treatments. The number of emerged panicles in C treatment was 3.0 and in D 11.9. However, 27.5 % of the largest tillers had initiated panicles also in treatment A at the end of the growing period, and the percentage of initiated tillers in the other treatments was 46.3 % (B), 65.0 % (C), and 93.8 % (D). The average length of the longest culm was only 1.2 cm in the treatment A compared to 3.1 cm in B, 55.2 cm in C, and 92.3 cm in D. The critical day-length was longer for culm elongation (12—14 h) than for floral initiation (about 10 h) in Haka cv.

Floral initiation was not detected to occur in the fall in field grown material. Only 1 per cent of the largest tillers had initiated panicles in the field plant samples on the 9th of February 1990. On the 26th of April 1990, a similar sample was taken, and most of the tillers studied was either at the transitional phase (50 %) or had already formed small (2—3 mm) panicles (42 %). 8 per cent of the largest tillers were still in the vegetative stage. The main period of floral initiation is the spring in the Finnish conditions.

Index words: *Dactylis glomerata*, orchard grass, induction, floral evocation, vernalization, initiation, winter, seed production, daylength.

In the seed production fields of cocksfoot in Finland the number of panicles has been usually very low (VALLE 1963, KÖYLIJÄRVI 1983). Therefore the average seed yield has remained low, too. In the variety trials of the Agricultural Research Centre the average seed yield has been only about 200 kg ha^{-1} (MUSTONEN et al. 1987) although also high yields from 600 to 870 kg ha^{-1} have been reported occasionally in some experiments (VALLE 1964, HAKKOLA 1967). The reasons for great variability and poor average panicle production are not clear. One possible reason may be inadequate floral evocation. Floral evocation is a complex process affected by many factors at different sequences and none of these factors alone can lead to flowering (BERNIER et al. 1981, BERNIER 1988). There are numerous reports on the floral evocation requirements of cocksfoot, however, the results are somewhat conflicting. Short daylength alone (CALDER 1963, 1964 b, WILSON and THOMAS 1971, BLONDON 1972, 1985, BROUE and NICHOLLS 1973, IKEGAYA et al. 1980) or low temperature alone (BLONDON 1972, 1985, IKEGAYA et al. 1980) has been reported to lead to floral evocation. According to GARDNER and LOOMIS (1953), HOVIN et al. (1966) and KOZUMPLIK and CHRISTIE (1972), both short daylength and low temperature are necessary for floral evocation. In the study of HEIDE (1987) with Scandinavian material floral evocation occurred in 8 h daylength at temperatures ranging from 9 to 21 °C. However, floral evocation occurred also in 24 h daylength at +3 °C temperature but evocation required more than 20 weeks. Probably the different genetic background (HOVIN et al. 1966, BROUE et al. 1967) and different age of the test plants (IKEGAYA et al. 1982) have led to conflicting results. The statement by BERNIER (1988), "Clearly , there are alternate pathways to flowering in all species (genotypes)", is true in the case of cocksfoot.

The stage of development of flower primordia before winter is an important factor for wintering ability since nearly all initiated tillers either died or were damaged during the winter in GARDNER and LOOMIS's (1953) experiment with cocksfoot, and similar results are reported concerning other perennial grass species, too (SASS and SKOGMAN 1951, ELLIOT 1966, HODGSON 1966). The floral evocation of cocksfoot has been observed to occur already in the fall but the transition from vegetative to generative growth has occurred in late winter (CANODE et al. 1972). However, in Northern conditions the initiation of inflorescences has been noticed to occur already in the fall in some perennial grass species (HODGSON 1966, HÅBJØRG 1979 a, b, HEIDE 1980, 1986, AITKEN 1985, HINTIKKA 1985). The possibility of cocksfoot to start floral initiation during short daylength has been observed by WYCHERLEY (1952) and BLONDON (1985), but HEIDE (1987), using Scandinavian material, reported that initiation of floral primordia did not take place in 8 h daylength but required a transition from short daylength to long daylength. Adaptation to daylength conditions in the fall is important with regards to both wintering ability and panicle formation ability (KLEBESADEL 1973).

The main aim of this study was to evaluate the fullfillment of the floral evocation requirement of cocksfoot in Finnish conditions. One purpose was to clarify to what extent cocksfoot is able to produce panicles in the fall before winter. Another target was to identify the negative or positive effect of winter on panicle formation ability. In addition, we tried to identify the time when the growth of cocksfoot changes from vegetative to generative phase in Finnish conditions.

Experimental design

To obtain sample plants for the trial a row seeded stand of cocksfoot was established at the Institute of Crop and Soil Science of Agricultural Research Centre of Finland in Jokioinen (latitude 60°49'N), located in Southern Finland. The Haka cultivar, released in 1981 by the Institute of Plant Breeding of the Finnish Agricultural Research Centre (RAVANTTI 1981), was used. Haka was seeded at a 50 row width on 24.6.1987 on a loam clay soil. A breeder's seed lot was used. Germination was 91 % and 1 000 seed weight 1.64 g. Sowing rate was 500 germinating seeds per m^2 (250 seeds per meter of a row). The nutrient content of the soil was (mg per 1 soil) Ca 2 950, K 260, Mg 540 and P 7.6. pH of the soil was 6.6. The basic fertilization at seeding was 500 kg mixed fertilizer (17-6-12 NPK) ha^{-1}. Actril S herbicide at the rate 3 1 ha^{-1} in 400 1 water was sprayed against weeds after emergence of the cocksfoot stand.

Sample plants were taken at two-week intervals in the fall and at possible times (when the soil was without a snow cover) during winter and spring in the winter seasons 1987/88 and 1988/89. A 15 cm long section of the row was sampled with a special sample shovel. In trial I (season 1987/88) 6 samples were taken per time and in trial II (season 1988/89) 10 samples were taken each time. The stand was managed like an ordinary seed production field during summer 1988.

Developmental stage of the growing point of main tillers of extra sample plants was analyzed with a microscope according to the method of JEATER (1956) when the sample plants were collected. Additional samples were taken in the fall 1987 at the last sampling date 1st of December 1987 and the plants were grown for 4 weeks at 10 h daylength at + 10 °C temperature. After that, the developmental stage of the growing points was analyzed.

The sample plants were planted in 5 liter plastic pots. The height of the pots was 19.5 cm and the diameter at the top 20.5 cm and at the bottom 17.0 cm. The soil of the sample filled about two-thirds of the pot and the pots were then filled up with a loam soil. Fertility values of the loam soil were: pH 5.7 and nutrients in mg per 1 soil: Ca 1334, K 410, Mg 86 and P 53. The plants were fertilized with 5 grams mixed fertilizer (17-6-12 NPK) per pot, and transfered into a glasshouse for growing. The level of fertilization was evaluated to be appropriate in the preliminary fertilization trials.

The growing period of the sample plants began immediately after taking the samples but if the soil was frozen during sampling, then samples were kept four days at + 5 °C temperature in natural light conditions to make the transition to warm conditions milder (see POHJAKALLIO et al. 1959 a, b). The plants were grown at + 18 °C temperature in continuous light.

Irradiation from the sodium lamps was about 80 Wm^{-2} at plant level (Li-1000, Li-Cor pyranometer sensor) without natural light. The amount of natural light was higher in the plants grown during the spring than in the plants grown in the fall due to longer daylength and higher light intensity. During bright spring days temperatures in the glasshouse occasionally increased above + 20 °C, sometimes to nearly 30 °C, because the fans could not maintain the set temperature.

The location of the plants in the glasshouse was according to randomized block design. The location of plants was so spacious that the plants sampled earlier did not shade those sampled later. The growing period in the glasshouse lasted for 10 weeks in trial I and 8 weeks in trial II. Most of the panicles were in the flowering phase when the trial was terminated. The date of panicle emergence was observed, and when the trial was terminated the number of panicles and the number of 'main

Table 1. Climatological data from June 1987 to June 1989 in Jokioinen. Data was obtained from the Jokioinen observatory (ANON. 1987—1989).

|	Monthly mean temperature	Long-range monthly mean temperature	Maximum temperature (date)	Minimum temperature (date)	Minimum temperature at soil/ snow surface (date)	Precipitation (mm)	Long-range average precipitation (mm)	Global radiation MJ/m²	Long-range average
1987									
June	12.1	13.7	23.3 (24)	4.6 (6)	0.7 (6)	81	42	416	(639)
July	14.8	16.2	28.4 (23)	2.2 (17)	−1.8 (17)	68	70	642	(566)
August	11.7	14.7	20.5 (21)	−0.1 (25)	−3.7 (25)	83	74	356	(445)
September	8.4	9.7	17.8 (6)	−2.2 (18)	−4.7 (18)	120	61	217	(246)
October	6.4	4.3	14.6 (2)	−2.2 (1)	−4.7 (1)	43	61	108	(108)
November	−0.7	−0.1	9.5 (26)	−13.2 (26)	−13.2 (26)	38	51	33	(31)
December	−5.3	−3.5	3.1 (2)	−24.7 (22)	−27.5 (22)	36	41	17	(15)
1988									
January	−3.1	−7.2	4.2 (3)	−21.4 (1)	−24.7 (1)	49	35	19	(28)
February	−4.3	−7.8	1.8 (13)	−21.0 (20)	−24.5 (20)	41	27	72	(92)
March	−3.5	−4.6	6.9 (31)	−19.3 (20)	−22.6 (20)	45	25	189	(252)
April	0.9	2.2	13.8 (29)	−13.4 (13)	−17.7 (13)	46	33	370	(394)
May	11.4	8.8	26.5 (29)	−3.7 (20)	−7.3 (20)	44	39	625	(563)
June	16.5	13.7	31.5 (28)	−0.8 (3)	−3.7 (3)	25	42	597	(639)
July	19.0	16.2	29.1 (18)	8.3 (24)	5.0 (24)	128	70	583	(566)
August	14.1	14.7	21.2 (15)	5.8 (29)	1.9 (29)	79	74	355	(445)
September	10.8	9.7	20.8 (3)	−2.7 (22)	−6.9 (22)	85	61	239	(246)
October	4.2	4.3	14.3 (6)	−13.2 (31)	−17.4 (31)	96	61	127	(108)
November	−3.9	−0.1	5.2 (11)	−22.4 (21)	−27.2 (21)	12	51	48	(31)
December	−7.0	−3.5	3.4 (29)	−24.5 (12)	−29.6 (12)	55	41	21	(15)
1989									
January	−0.5	−7.2	4.3 (17)	−18.4 (2)	−20.8 (2)	33	35	30	(28)
February	0.0	−7.8	7.2 (3)	−12.7 (18)	−17.7 (18)	61	27	78	(92)
March	1.1	−4.6	7.9 (21)	−5.7 (31)	−7.8 (31)	40	25	141	(252)
April	5.3	2.2	20.4 (13)	−9.6 (4)	−12.2 (4)	40	33	343	(394)
May	10.4	8.8	25.8 (24)	−1.6 (1)	−5.0 (12)	41	39	617	(563)
June	15.4	13.7	26.8 (28)	4.8 (14)	0.8 (2)	30	42	636	(639)

Fig. 1. Daily values for thickness of snow cover (——) and soil temperature at 1 cm depth (·—·—) in grass field on clay soil at the Jokioinen observatory during the winter season 1987/1988.

tillers' (diameter at the base of tiller > 5 mm) were counted per pot. The dry weight of panicles and the dry weight of the above ground phytomass was measured.

Critical daylength for panicle initiation and for culm elongation of Haka cv. was studied in a separate trial. The production of the experimental material was similar to that earlier mentioned. Samples were taken on the 9th of February in 1990 when the snow had melted and the soil thawed. Natural daylength is 10 h at that time of the year in Jokioinen. Eight samples were taken for each treatment.

The stage of development of growing point of 100 main tillers was analyzed on the 9th of February when the sample plants were taken from the field.

The daylength treatments were performed in growth chambers at constant + 15 °C temperature. The daylength treatments were A) 10 h, B) 12 h, C) 14 h, and D) 16 h. The irradiation in all treatments was 105 Wm^{-2} for the first 10

hour photoperiod. For the rest of the photoperiod in treatments B, C, and D the irradiation was 20 Wm^{-2}.

The trial was terminated after a 9 week growing period. The number of emerged panicles was counted per pot, and the length of the longest leave per pot measured as well the total above ground phytomass. Additionally, the growing point of the 10 largest tillers of each pot was analyzed, and the length of culms measured. If the growing point was at the vegetative stage culm length was marked to be 0 mm. The developmental stage of the growing point was divided into three categories: vegetative, transitional, or generative.

Sample plants were taken also on the 26th of April 1990 from the same field grown stand to detect the time of the initiation of panicles in natural conditions. The development stage of the growing point of 100 main tillers was analyzed.

The data was subjected to the analysis of var-

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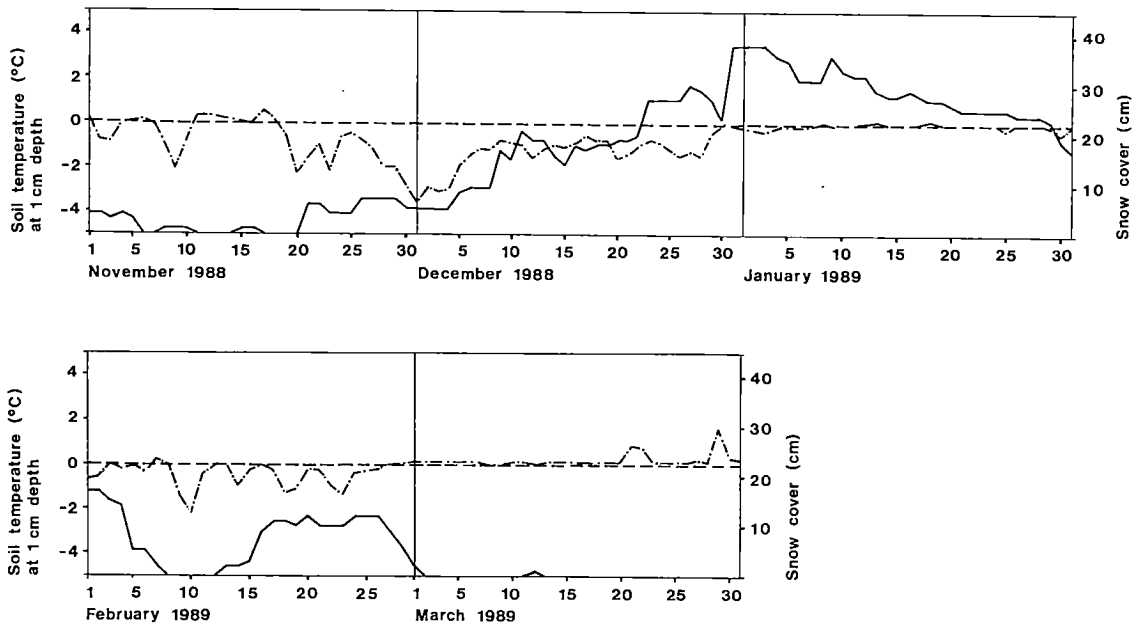
Fig. 2. Daily values for thickness of snow cover (———) and soil temperature at 1 cm depth (·—·—) in grass field on clay soil at the Jokioinen observatory during the winter season 1988/1989.
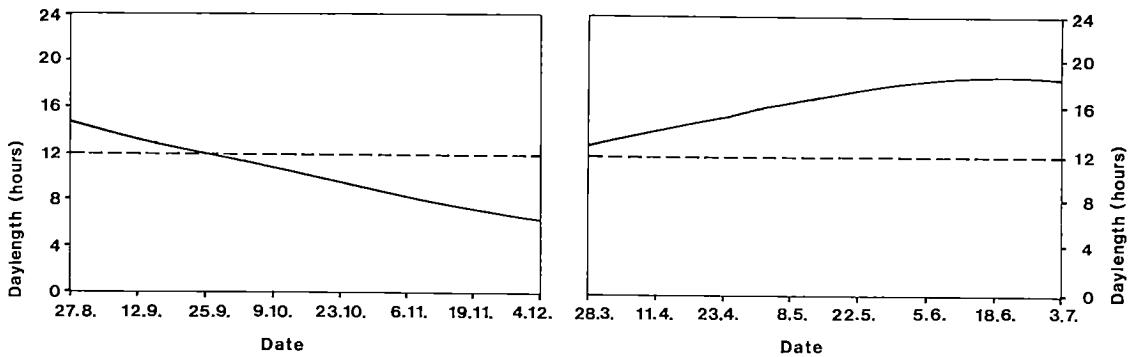
Fig. 3. Daylength in Jokioinen in fall and spring according to sunrise and sunset times.

iance according to the randomized block design in the SPSS^x statistical package (ANON. 1986). The significance of F values is expressed as follows: *** = P > 0.999, ** = P > 0.99, * = P > 0.95.

The significance of the difference of means was tested with the Tukey's multiple comparison test (SNEDECOR and COCHRAN 1978) at the risk level P < 0.05.

Weather conditions

Weather data was obtained from the Jokioinen observatory (ANON. 1987—1989) located about 1 000 m from the experimental field. Meteorological measurements were taken from field conditions similar to those of the experimental field.

The monthly minimum, maximum and average mean temperatures of the weather cage at

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2 m height between June 1987 and June 1989 are presented in Table 1. The monthly precipitation is also presented in Table 1. Thickness of snow cover and the temperature conditions at 1 cm depth of soil during the winter seasons 1987/88 and 1988/89 are expressed in Figs. 1 and 2. The daylength in the fall and in the spring in Jokioinen is shown in Fig. 3.

RESULTS

The plants which were transferred into the glasshouse early in the fall did not form panicles at all (Tables 2 and 3). The ability to produce panicles developed gradually, but in both trials also plants sampled before winter were able to produce panicles. Panicle formation ability was therefore already present in the fall but strengthened during the winter. Ability to produce panicles increased in both trials from fall to spring. Sampling dates and the values of the number of panicles and 'main tillers' and the dry weight of panicles and above ground phytomass are presented in Tables 2 and 3.

The growing point of tillers remained at the

Table 2. Number of panicles, dry weight of panicles and of above ground phytomass, and number of main tillers at different times in 1987 and 1988 of cocksfoot cv. Haka sample stands transferred from field into glasshouse. Favourable conditions for panicle growth lasted 8 weeks. n = 6. Values per pot.

Date of transfer	Number of panicles	Dry weight of panicles (g)	Dry weight of above ground phytomass (g)	Number of main tillers
07.09.1987	0[e]	0[c]	73.1[bcd]	36.0[cd]
21.09.1987	0[e]	0[c]	75.1[bcd]	32.5[cd]
05.10.1987	0.2[e]	0.1[c]	87.7[abc]	39.4[bc]
19.10.1987	0.8[e]	0.3[c]	86.8[abc]	49.8[ab]
02.11.1987	4.2[de]	1.0[bc]	112.5[a]	38.7[b]
16.11.1987	1.0[e]	0.3[c]	91.1[abc]	24.7[e]
01.12.1987	4.7[cde]	1.4[bc]	109.2[a]	33.9[cd]
04.01.1988	12.5[bcd]	4.7[ab]	103.2[ab]	44.8[ab]
20.04.1988	13.3[abc]	3.0[b]	50.3[d]	44.0[ab]
04.05.1988	20.5[ab]	5.8[a]	52.5[d]	45.8[ab]
19.05.1988	22.0[a]	3.3[b]	62.6[cd]	30.8[de]

[a–e] Means with a different superscript letter within a column are significantly different (P<0.05).

Table 3. Number of panicles, dry weight of panicles and of above ground phytomass, and number of main tillers at different times in 1988 and 1989 of cocksfoot cv. Haka sample stands transferred from field into glasshouse. Favourable conditions for panicle growth lasted 8 weeks. n = 10. Values per pot.

Date of transfer	Number of panicles	Dry weight of panicles (g)	Dry weight of above ground phytomass (g)	Number of main tillers
15.09.1988	0.1[e]	0.0[d]	78.8[a]	28.9[ab]
29.09.1988	1.5[de]	0.5[cd]	66.8[ab]	31.4[a]
13.10.1988	3.9[cde]	1.0[cd]	56.7[b]	22.4[ac]
10.11.1988	3.9[cde]	1.0[cd]	61.4[ab]	21.2[bc]
24.11.1988	5.6[bde]	1.3[bd]	57.5[b]	18.0[c]
07.02.1989	9.3[bc]	2.5[bc]	51.6[b]	23.6[ac]
14.03.1989	8.4[bd]	2.6[bc]	50.5[b]	24.8[ac]
19.04.1989	12.2[ab]	3.3[b]	52.6[b]	27.2[ac]
09.05.1989	18.2[a]	5.9[a]	49.4[b]	29.7[ab]

[a–e] See table 2.

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Table 4. Effect of daylength on floral initiation, culm length, emergence of panicles, length of leaves, and on above ground phytomass production of cocksfoot. Flower induced cocksfoot plants were grown 9 weeks at 10, 12, 14 or 16 hour daylength at $+15\ ^{\circ}C$ temperature. Irradiation was 105 Wm^{-2} for the first 10 hours and 20 Wm^{-2} for the rest of photoperiod. n = 8. Values per pot.

Daylength	Number of emerged panicles	Initiated tillers %[x]	Length of culm[y] leaves[z] (cm)		Dry weight of above ground phytomass (g)
A. 10 hours	0.0[b]	27.5[c]	1.2[c]	78[b]	21.4[c]
B. 12 hours	0.0[b]	46.3[bc]	3.1[bc]	108[a]	32.4[ab]
C. 14 hours	3.0[b]	65.0[ab]	55.2[b]	110[a]	39.0[a]
D. 16 hours	11.9[a]	93.8[a]	92.3[a]	107[a]	27.9[b]

[a-c] See Table 2.
[x]   Average of the 10 most advanced tillers in each pot.
[y]   Average of the longest culm in each pot.
[z]   Average of the longest leave in each pot.

vegetative stage in this material at all sampling dates in the fall and winter. Only on the last sampling times in the spring were the growing points differentiated to flower primordia. The growing point of those additional sample plants taken on the 1st of December 1987 which were kept four weeks at 10 h daylength at 10 °C temperature remained also at the vegetative stage.

In the daylength trial 99 of the 100 main tillers analyzed were in the vegetative stage at the beginning of the treatment period 9th of February 1990. Only 1 panicle was initiated. The length of the panicle was 2 mm. No panicles emerged during the 9 weeks growing period in the 10 and 12 hour treatments (Table 4). Significantly more panicles emerged by the treatment at 16 h compared with the other treatments. However, some panicles had initiated also in the shorter daylengths (Table 4) but because of the slight elongation of the culms the panicles did not emerge. The percentage of initiated tillers was significantly higher in the 16 h and 14 h daylength treatments than in the 10 hour treatment.

From the field samples taken on the 26th of April most of the growing points of the tillers were at the transitional phase (50 %) or had already formed small panicles (42 %). Eight per cent of the large tillers studied had remained at the vegetative stage. Daylength is about 14 hours at the end of April in Jokioinen.

DISCUSSION

Features of the experimental design

The fact that the samples taken on different dates were subjected to slightly different environmental conditions during the growing period in the glasshouse causes one source of variation in this trial. On the one hand, the natural irradiation was considerably higher in spring grown samples than in the fall grown samples. On the other hand, the temperature increased occasionally above + 18 °C in the spring when the outdoor air temperature had also risen so high that the fans could not maintain the set temperature.

Reduced light intensity before spikelet initiation decreased the number of ear-bearing tillers in fescue in RYLE's (1967) trial. The greater production of above ground phytomass in the fall than in the spring grown sample plants in the present study indicates that light inten-

sity was sufficient also in the fall for the growth of cocksfoot. The amount of reserve carbohydrates per plant is greater in the fall than in spring, and that may have positively influenced the production of phytomass in fall samples compared to spring samples.

The occasional high temperature in the spring was presumably of no advantage to panicle production. According to HANSON and SPRAGUE (1953), CALDER (1964 a), and BROUE and NICHOLLS (1973) high temperatures inhibit the initiation and growth of panicles. However, BLONDON (1985) suggested that the most favourable temperature for floral development varies between 17 and 33 °C, according to the genotypes.

In spite of the rather late sowing date (24.6.1987) of the stand the effect of the juvenile stage of cocksfoot (COOPER and CALDER 1964, IKEGAYA et al. 1981, HEIDE 1987) probably did not affect the results of Trial I. The number of panicles produced per sample in the spring in Trial I was as high as the highest number in the experiment on juvenility in 1988 (NIEMELÄINEN 1990 a) thus the effect of juvenility cannot be strong.

The 15 cm long samples from the row of seeds established stand of a crosspollinated species is always more or less heterogeneous genetically. The number of 'main tillers' was counted to roughly evaluate the eveness of the structure of the samples. Although there are significant differences in the number of 'main tillers' between different sampling dates, the variation in the number of 'main tillers' is small compared to the variation in the number of panicles. Therefore, it is assumed that the variation in the samples did not affect greatly the most important character, the number of panicles per sample, in the study.

Initiation of floral primordia

The initiation of flower primordia in the fall was not observed in the present material. This sup-ports the findings from southern latitudes (BOMMER 1959, ANON. 1978) that the transition from vegetative growth to generative growth occurs in the spring in cocksfoot. The result does not, on the other hand, refute the findings of WYCHERLEY (1952) and CALDER (1964 b) that the flower initiation of cocksfoot can already begin during the short days in fall. In fact, the relatively high percentage of initiated panicles in the most advanced tillers in the 10 h (27.5 %) and 12 h (46.3 %) daylength treatments indicates that the initiation of cocksfoot is possible already in the fall in the Finnish material as well. The 9 week growing period at + 15 °C is, however, improbable in Finnish conditions in the fall after flower evocation requirements are met. The additional test plants grown in the fall in Trial I for 4 weeks in 10 h daylength at + 10 °C without any sign of floral initiation supports the conclusion that the occurrence of floral initiation in the fall is improbable.

The critical daylength was about 10 h for panicle initiation and between 12 and 14 hours for culm elongation in the present material. This supports the findings of CALDER (1964 b) and WILSON and THOMAS (1971) with cocksfoot and the finding of HÅBJØRG (1978) with Kentucky bluegrass that floral initiation and culm elongation have a different critical daylength. According to CALDER (1964 b) the floral initiation of cocksfoot can occur already at short daylength but culm elongation does not occur before daylength is 11—12 h or longer. In the study of WILSON and THOMAS (1971) in New Zealand, initiation occurred at 11 h daylength and panicle emergence occurred at 14 h daylength. In the study of HEIDE (1987) on Scandinavian cultivars, no stem elongation occurred at 8 h photoperiod and only a few cm elongation occurred in 13 h photoperiod at + 15 °C in a 4-week treatment period.

In field grown plants the transition from vegetative growth to generative growth occurs mainly in the spring. Although the probability

of floral initiation in the fall is slight in Finnish conditions, it may occur in most advanced stands (NIEMELÄINEN 1990 a), and the phenomenon needs further study especially in those cases when the stand is established early in spring without a cover crop. HEIDE (1990) stresses the great variation in the critical daylength and temperature conditions for evocation and especially in the duration of evocative period between the ecotypes within each species.

The rate of development was most rapid in the longer daylength treatment similarly as in the trials of GARDNER and LOOMIS (1953) and CALDER (1964 a). However, when the critical daylength is passed, temperature is the main factor causing variation in the rate of development (HANSON and SPRAGUE 1953, BEDDOWS 1968).

Development of panicle production ability

Panicle production ability was already present in the fall as in CANODE's et al. (1972) trial. However, the panicle production ability increased significantly during the winter and spring. According to BLONDON (1985) temperatures between $+2$ and $+10$ °C cause floral evocation in cocksfoot. HEIDE (1987) revealed that floral evocation in Scandinavian cocksfoot material occurs also in long photoperiod when the temperature is lowered to $+3$ °C. During the winter, the temperature is occasionally favourable for floral evocation although during most of the winter the temperature is below $+2$ °C around the growing point tissue (Figs. 1 and 2). It is unclear if plants that are dormant in the winter can be vernalized because dividing cells should be present at vernalization. The growth and metabolism of cocksfoot was observed between 0 and $+2$ °C temperature in the trial of OBRAZTSOV et al. (1980) but the growth of leaves ceased already at $+3$ °C temperature in DAVIDSON and MILTHORPE's (1965) trial. The present data suggests that conditions

in March and in April were favourable for floral evocation and increased the panicle production ability. According to BERNIER (1988) the summation of different inductive factors is an important feature of the floral evocation phenomenon.

Effect of climatological conditions

The climatological conditions during the winter season 1987/88 were such that no considerable risk for panicle formation was obvious. The temperature was so high in the fall before snow cover came that the risk of freezing injury was low. The lowest temperature (-24.7 °C) occurred when the snow cover was so thick that the temperature at soil surface did not fall below -2.5 °C. The protective effect of snow (YLIMÄKI 1962, NIEMELÄINEN 1990 b) was obvious in this trial. In January 1988 occurred the common situation in Southern Finland that a mild weather period melted the snow cover completely. Fortunately, the temperature remained mild during the snowless period. The snow cover lasted until the middle of April and the temperatures in April were so mild that no freezing injury occurred. Although cocksfoot is sensitive to freezing damage, compared to many other perennial grasses, cocksfoot tolerated temperatures down to -18 degrees Celcius in the trials of LIMIN and FOWLER (1987). In the trials of GUDLEIFSSON et al. (1986) 50 per cent of shoots of cocksfoot trimmed to 3 cm tolerated temperatures down to -8.5 °C.

During the winter season 1988/89 the protective effect of snow cover was also obvious. Before a snow cover was formed, the lowest temperature was -13 °C in November. Immediately after the snow cover had formed the temperature fell to -22.4 °C. The snow cover was already 20 cm thick when a -29.4 °C frost occurred on 12th of December in 1988. In spite of so low a temperature in the weather cage at a height of 2 meters the temperature at 1 cm depth in the soil did not fall below -4 °C dur-

ing the whole winter. Already, quite a shallow snow cover with the upward flux of heat from the soil kept the temperature at the surface of the soil safe for the growing points. The snow melted completely in February in 1989 but the period was short and the weather remained mild during the snowless time. The snow cover disappeared already by the 2nd of March in 1989, but the temperature was so high in the spring that the risk of freezing injury was small. The plants survived those fall frosts well which occurred before snow cover was formed (minimum temperature -13 °C). According to GUSTA (1986) the freezing tolerance of plants is highest in early winter but decreases rapidly in the spring.

The present material is not sufficient to precisely evaluate the influence of the winter on panicle production of cocksfoot. Cocksfoot attained the ability to produce panicles already in the fall. Panicle production ability in the fall was, however, so small that for abundant panicle production the conditions during winter and spring have to be positive. The effect of winter can be positive to panicle formation, as in this study, but the present study does not exclude the possibility that during some conditions the winter can affect negatively the panicle formation (LÜTKE-ENTRUP and SCHRIMPF 1964, NIEMELÄINEN 1990 b).

Practical considerations

In the present study the number of panicles has been used to evaluate the seed production properties of cocksfoot. The correlation coefficient between the number of panicles per m² and seed yield of cocksfoot varied from 0.62 to 0.88 in different years and was statistically significant in every year studied in the studies of NORDESTGAARD and LARSEN (1974). However, one has to be cautious when the results of this pot trial are applied to practical seed production conditions in the field. Transformation of number of panicles per pot to the number of panicles per m² is not easy because the chosen row width of a stand strongly affects the value. The ordinary row width in the seed production of cocksfoot is 12.5 cm, but the growing of sample plants in the glasshouse was so spacious that so narrow a row width is not granted. When the 50 cm row width is used the maximal calculated number of panicles per m² was 290 in May 1988 and 240 in May 1989. The values calculated according to 50 cm row width are probably low than high estimates.

The calculated number of panicles per m² is quite low compared to the numbers reported from Denmark where the number of panicles per m² has ranged from 430 to 950 in various treatments and field experiments (NORDESTGAARD 1979 and 1981). The seed yield in those experiments varied from 510 to 1 270 kg ha^{-1}. In Finland the number of panicles per m² have ranged from just a few to up to 600 in different field trials and treatments (NIEMELÄINEN, unpubl.). The harvested seed yield varied between 0 to 900 kg ha^{-1} in those trials.

Although in this trial the per m² calculated panicle production of sample stands was not high, the floral evocation requirements of cocksfoot were met to such an extent in the spring that inadequate floral evocation can not be the primary reason for poor panicle production in Finnish conditions when the stand is in good condition in the fall, and when conditions for the growth of panicles are favourable.

Because flower initiation in the fall was not detected in this material, the risk of injury to initiated inflorecences during winter is small.

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SELOSTUS

Koiranheinän (Dactylis glomerata L.) röyhynmuodostukseen vaikuttavista tekijöistä Suomessa.

I. Röyhynmuodostuskyvyn kehittyminen ja kukintojen kasvun alkamisen ajankohta Jokioisissa

OIVA NIEMELÄINEN

Maatalouden tutkimuskeskus

Koiranheinän röyhynmuodostus on ollut maamme siemenviljelyksillä hyvin vaihtelevaa. Usein röyhyjä on muodostunut vain vähän vaikka kasvusto on tuottanut runsaasti vihermassaa. Vähäinen röyhynmuodostus on johtanut pieneen hehtaarisatoon. Pienestä hehtaarisadosta johtuen maassamme ei ole koiranheinän siemenviljelyä siementarvetta vastaavasti.

Maatalouden tutkimuskeskuksen kasvintuotannon tutkimuslaitoksella selvitettiin syitä koiranheinän heikkoon röyhynmuodostukseen tutkimalla eri tekijöiden vaikutusta astiakokeissa kasvihuoneessa ja/tai kasvatuskammioissa. Tavoitteena oli yksilöityjen koejärjestelyjen avulla selvittää mitkä tekijät olennaisimmin vaikuttavat röyhynmuodostuksen epäonnistumiseen Suomessa. Tässä artikkelissa selvitetään

kukinnan virittymisen vaatimusten täyttymistä ja röyhyn-muodostuskyvyn kehittymistä syksyn, talven ja kevään aikana. Koiranheinän röyhynkasvun alkamisen ajankohtaa tarkastellaan siltä kannalta alkaako röyhynkasvu jo ennen talvea, jolloin talven haitalliset olosuhteet voisivat erityisesti vaikuttaa röyhyjen aiheisiin.

Jokioisissa eri aikoina syksyllä, talvella ja keväällä syksyn 1987 ja kevään 1989 välisenä aikana kentältä kasvihuoneeseen siirrettyjen Haka-lajikkeen näytekasvustojen röyhyn-muodostus oli pienin syyskuussa (0—1,5 röyhyä/astia), ja suurin huhtikuussa (12,2—13,3) ja toukokuussa (18,2—22,0) otetuissa näytteissä. Ennen joulukuuta otetut näytekasvustot tuottivat vain vähän röyhyjä. Röyhyjen muodostuskyky oli kuitenkin olemassa pienessä määrin jo syksyllä, mutta se lisääntyi talven ja kevään aikana. Röyhynmuodostuskyky oli syksyllä niin vähäinen, että talven ja kevään olosuhteiden tulee olla röyhynmuodostukselle edulliset jotta kaupallinen siemenviljely olisi mahdollista.

Erillisessä kokeessa tutkittiin kuinka pitkä päivänpituuden tulee olla ennen kuin kukkimaan virittyneen koiranheinän kukintojen kehitys alkaa. Pellolta 9. helmikuuta 1990 otettuja näytekasveja kasvatettiin 10, 12, 14 ja 16 tunnin päivänpituudessa + 15 asteen lämpötilassa 9 viikkoa ja röyhynmuodostus ja varren pituuskasvu määritettiin.

Kymmenen ja 12 tunnin käsittelyissä röyhyjä ei tullut lainkaan esiin kun sen sijaan 14 ja 16 tunnin päivänpituudessa esiintulleiden röyhyjen lukumäärä oli keskimäärin 3,0 ja 11,9. Kuitenkin 27,5 % myös 10 tunnin päivänpituudessa

kasvaneista suurimpien versojen kasvupisteistä oli jo erilaistunut röyhyksi. Vastaava osuus muissa päivänpituuksissa oli 46,3 % (12 h), 65,0 % (14 h) ja 93,8 (16 h). Pisimmän korren pituus eri käsittelyissä oli keskimäärin 1,2 cm (10 h), 3,1 cm (12 h), 55,2 cm (14 h), ja 92,3 cm (16 h). Kriittinen päivänpituus oli pienempi röyhyn kehityksen alkamiselle (10—12 h) kuin varren pituuskasvun alkamiselle (> 12 h). Tulosten perusteella kukkimaan virittyneen Haka-koiranheinän versot voivat teoriassa aloittaa röyhyjen kasvun jo syksyn päivänpituuksissa, mutta tarvittavan pitkän lämpimän kauden esiintyminen on epätodennäköistä.

Kokeessa, jossa näytekasveja otettiin kentältä eri aikaan syksyllä, talvella ja keväällä röyhyjen kasvun alkamista ei havaittu ennen kuin huhtikuussa. Pääasiallinen röyhyjen kasvun alkamisen ajankohta Suomen olosuhteissa on kevät.

Suurin astiakoetuloksista laskettu koiranheinän röyhyn-muodostus neliömetriä kohti oli 290 röyhyä toukokuussa 1988 ja 240 röyhyä m^{-2} toukokuussa 1989. Vaikka röyhyjen muodostus pinta-alaa kohti oli varsin pieni oli se molempina vuosina niin runsas, että epätäydellinen koiranheinän kukinnan virittyminen ei ole pääasiallinen syy koiranheinän vähäiseen röyhynmuodostukseen silloin kun kasvusto on syksyllä hyvässä kunnossa ja olosuhteet ovat keväällä suotuisat röyhyjen kasvulle ja kehitykselle. Koska röyhyjen kasvun alkamista syksyllä ei havaittu, vähentää tämä talven alhaisten lämpötilojen aiheuttamaa riskiä röyhyjen kasvulle.

FACTORS AFFECTING PANICLE PRODUCTION OF COCKSFOOT (DACTYLIS GLOMERATA L.) IN FINLAND

II. EFFECT OF JUVENILE PHASE, SOWING DATE, AND TILLERING

OIVA NIEMELÄINEN

NIEMELÄINEN, O. 1990. Factors affecting panicle production of cocksfoot *(Dactylis glomerata* L.) in Finland. II. Effect of juvenile phase, sowing date, and tillering. Ann. Agric. Fenn. 29: 231—239. (Agric. Res. Centre, SF-31600 Jokioinen, Finland.)

Cocksfoot cv. Haka was sown in a row stand at two-week intervals between 18.5.1988 and 10.8.1988 in Jokioinen. The panicle production of sample plants was studied before and after winter 1988/89 by growing the sample plants in a glasshouse in favourable conditions for panicle growth.

The production of panicles per sample was significantly higher for the first two sowing dates (16.1—21.6) than at later sowing dates (0.1—8.1). In the field grown stand the first two sowing dates produced significantly more panicles per meter of row than the later sowing times, too. An early sowing date was necessary for the abundant panicle production of cocksfoot in the first harvest year.

In a separate pot trial a cocksfoot stand was established from one seed at two week-intervals between 6.6.1989 and 1.8.1989. Panicle production was measured in the fall 1989. All three first sowing dates produced a high number of panicles per pot (35.8—41.3) which indicates that the lateral tillers contribute greatly to the panicle production of cocksfoot. The stand of the last sowing date produced significantly less panicles per pot (11.8) than the earlier sowing dates.

Floral initiation was observed to occur in the most advanced tillers already in the fall in the pot seeded trial.

Index words: *Dactylis glomerata,* orchard grass, seed production, establishment.

INTRODUCTION

Most grasses grow vegetatively some time after sowing although the conditions for flowering are ideal then. This stage of vegetative growth is called the juvenile stage. When a plant is in the juvenile stage, vegetative growth continues and the plant is not evoked to flower in conditions which later on evoke flowering (BERNIER et al. 1981).

The juvenile stage of cocksfoot has been detected in the studies of CALDER 1963 and 1964 a, COOPER and CALDER 1964, CANODE et al. 1972, BROUE 1973, HEIDE 1987. The duration of the juvenile phase has been estimated to last from 4 to 7 weeks from sowing (CALDER 1963 and 1964 a, IKEGAYA et al. 1979 and 1981 a, HEIDE 1987), or until a certain amount of leaves have developed on the plant (KOZUMPLIK and CHRISTIE 1972 a, BROUE 1973, IKEGAYA et al. 1981 a, b, BLONDON 1985). In different studies, the juvenile stage has been observed to be

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passed when 5 to 12 leaves have developed.

In the present study the effect of juvenile stage on the panicle formation ability of cocksfoot was studied by using different sowing dates. The different growing time of the sample stands, before inductive conditions began, resulted in variable amount of tillering, and the effect of tillering on panicle production is discussed. The aim of the study was to evaluate the role of the juvenile phase on the observed poor panicle and seed production of cocksfoot in the stands on the first seed production year in Finland.

MATERIAL AND METHODS

Results are presented from two separate trials. In Trial I field grown sample stands were used. In Trial II seeds were sown directly in pots. In Trial I Haka cultivar was sown at two-week intervals from 18.5.1988 to 10.8.1988 on loam clay soil at 50 cm row width at the Institute of Crop and Soil Science in Jokioinen. Sowing rate was 500 germinating seeds per m². The whole experimental field was fertilized with 300 kg mixed fertilizer (17-6-12 NPK) before the first sowing time. Nutrient content of the soil was (mg in l of soil): P 11.0, K 310, Ca 2050 and Mg 335. pH was 5.7.

Weeds were sprayed with 3 l Actril S herbicide in 400 l water ha^{-1} after the emergence of the first two sowings. In trial I the sample plants were taken both prior to (24.11.1988) and after winter (9.5.1989). A 15 cm long section of row was taken with a sample shovel and the sample was planted in a 5 liter plastic pot. In addition, samples were taken also from a row stand which was sown on the 24th of June in 1987, and was managed in 1988 as a seed production field. The stand was fertilized with 300 kg mixed fertilizer (17-6-12 NPK) after the seed harvest. The pots were filled up with loam soil and the sample plants fertilized with 5 g mixed fertilizer (17-6-12 NPK) per pot. Ten samples were taken of each sowing date. The plants were transferred into a glasshouse. In the fall sample plants were kept 4 days at + 5 °C in natural light conditions before the start of the growing period. Growing was conducted in continuous light at about + 18 °C temperature. Location of the plants in the glasshouse was according to the randomized block design.

The number of panicles was counted also in the field stand in the summer 1989 from a 50 cm piece of row at 10 places of each sowing time. Because of the uneven emergence of stand in sowing dates 3 and 4, the sample areas were chosen so that the stand was even without patches in the sample places. In the spring the field stand was fertilized with 600 kg mixed fertilizer (17-6-12 NPK) after the sample plants were taken into the glasshouse.

In Trial II, two seeds were sown in 5 l plastic pots at two-week intervals between 6.6.1989 and 1.8.1989. Loam soil was used in the pots. The pots were fertilized at the rate of 5 g mixed fertilizer (17-6-12 NPK) at sowing and again similarly at the start of the growing period in the glasshouse. The seedlings were thinned so that the stand originated from one seed. The pots were watered when necessary and were kept under natural daylight and temperature conditions until 24 November 1989 when the growing period in the glasshouse started.

In both trials, the number of panicles and the number of main tillers (diameter at the base of stem > 5 mm) were counted and the dry weight of panicles and above ground phytomass measured at the termination of the trial. In Trial II the number of main tillers was counted also at the beginning of the growing period. In the

Table 1. Climatological data from May 1988 to December 1989 in Jokioinen. Data was obtained from the Jokioinen observatory (ANON. 1988—1989).

	Monthly mean temperature	Long-range monthly mean temperature	Maximum temperature (date)	Minimum temperature (date)	Minimum temperature at soil/snow surface (date)	Precipitation (mm)	Long-range average precipitation (mm)	Global radiation MJ/m²	Long-range average
1988									
May	11.4	8.8	26.5 (29)	-3.7 (20)	-7.3 (20)	44	39	625	(563)
June	16.5	13.7	31.5 (28)	-0.8 (3)	-3.7 (3)	25	42	597	(639)
July	19.0	16.2	29.1 (18)	8.3 (24)	5.0 (24)	128	70	583	(566)
August	14.1	14.7	21.2 (15)	5.8 (29)	1.9 (29)	79	74	355	(445)
September	10.8	9.7	20.8 (3)	-2.7 (22)	-6.9 (22)	85	61	239	(246)
October	4.2	4.3	14.3 (6)	-13.2 (31)	-17.4 (31)	96	61	127	(108)
November	-3.9	-0.1	5.2 (11)	-22.4 (21)	-27.2 (21)	12	51	48	(31)
December	-7.0	-3.5	3.4 (29)	-24.5 (12)	-29.6 (12)	55	41	21	(15)
1989									
January	-0.5	-7.2	4.3 (17)	-18.4 (2)	-20.8 (2)	33	35	30	(28)
February	0.0	-7.8	7.2 (3)	-12.7 (18)	-17.7 (18)	61	27	78	(92)
March	1.1	-4.6	7.9 (21)	-5.7 (31)	-7.8 (31)	40	25	141	(252)
April	5.3	2.2	20.4 (13)	-9.6 (4)	-12.2 (4)	40	33	343	(394)
May	10.4	8.8	25.8 (24)	-1.6 (1)	-5.0 (12)	41	39	617	(563)
June	15.4	13.7	26.8 (28)	4.8 (14)	0.8 (2)	30	42	636	(639)
July	16.3	16.2	29.9 (8)	5.2 (21)	3.7 (21)	85	70	594	(566)
August	13.7	14.7	25.8 (3)	0.5 (31)	-2.7 (31)	92	74	368	(445)
September	11.0	9.7	20.0 (23)	-1.9 (12)	-5.0 (12)	50	61	274	(246)
October	4.7	4.3	11.5 (4)	-4.3 (7)	-8.2 (7)	49	61	113	(108)
November	-0.1	-0.1	9.0 (12)	-24.5 (27)	-28.2 (27)	69	51	31	(31)
December	-5.9	-3.5	4.3 (3)	-26.4 (16)	-30.9 (18)	47	41	19	(15)

field grown stand only the number of panicles was counted.

The data was subjected to the analysis of variance, and the significance of the difference of the means was tested using Tukey's multiple comparison -test (SNEDECOR and COCHRAN 1978). The climatological conditions in summer and fall in 1988 and 1989 are presented in Table 1. The length of day and the wintering conditions during the winter season 1988/89 in Jokioinen are presented in the paper of NIEMELÄINEN (1990).

RESULTS

In Trial I, the size of sample plants at the beginning of growing period was larger in sowings 1 and 2, and smaller in sowings 6 and 7 than in other sowings. However, the stands sown at different dates did not form a clear series of stands at the progressive stage of development. Especially the stands of sowing dates 3, 4 and 5 in Trial I were quite similar in the fall. The reason for this similarity was the dry early summer in 1988 which caused a slow and uneven emergence of the stands in sowing times 3 and 4. The plants from the overwintered 1987 sown stand differed from the stands sown in 1988 because the overwintered stand

Table 2. Growth and panicle production of cocksfoot sown in the field at two-week intervals beginning 18.5.1988 and after subsequent growing in a glasshouse. The samples were taken into the glasshouse 24.11.1988. n = 10. Values per pot.

Sowing date in 1988	Number of panicles	Dry weight (g) of panicles	Dry weight (g) of above ground phytomass	Number of main tillers
0) 24.6.1987*	7.4[b]	2.3[b]	62.7[b]	19.1[a]
1) 18.5.1988	17.0[a]	4.9[a]	83.4[a]	21.3[a]
2) 01.6.1988	19.6[a]	5.2[a]	74.9[ab]	16.8[a]
3) 15.6.1988	0.7[c]	0.3[c]	43.6[c]	21.2[a]
4) 29.6.1988	0.9[bc]	0.3[c]	45.3[c]	22.8[a]
5) 13.7.1988	2.4[bc]	1.0[bc]	49.8[c]	26.1[a]
6) 27.7.1988	0.6[c]	0.3[c]	44.1[c]	20.9[a]
7) 10.8.1988	0.1[c]	0.0[c]	44.5[c]	19.7[a]

[a—c] Means with a different superscript letter within a column are significantly different ($P < 0.05$).

* The stand was sown in 1987 and was managed as a seed production stand in 1988.

Table 3. Growth and panicle production of cocksfoot sown in the field at two-week intervals beginning 18.5.1988 and after subsequent growing in a glasshouse. The samples were taken into the glasshouse 9.5.1989. n = 10. Values per pot.

Sowing date in 1988	Number of panicles	Dry weight (g) of panicles	Dry weight (g) of above ground phytomass	Number of main tillers
0) 24.6.1987*	18.2[a]	5.9[a]	49.4[ab]	29.7[ab]
1) 18.5.1988	16.1[a]	4.9[a]	53.4[a]	32.9[a]
2) 01.6.1988	21.6[a]	6.7[a]	55.8[a]	34.8[a]
3) 15.6.1988	2.1[b]	1.0[b]	35.6[c]	23.9[bc]
4) 29.6.1988	8.1[b]	2.0[b]	35.7[c]	23.8[bc]
5) 13.7.1988	5.9[b]	1.5[b]	34.3[c]	20.9[c]
6) 27.7.1988	8.1[b]	2.5[b]	42.0[b]	24.0[bc]
7) 10.8.1988	2.0[b]	0.8[b]	33.7[c]	18.6[c]

[a—c] See table 2.

* The stand was sown in 1987 and was managed as a seed production stand in 1988.

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Table 4. Effect of sowing date on the number of panicles and on the dry weight of panicles in the field grown cocksfoot stand in 1989. Panicles were counted and measured from a 50 cm long piece of a row. Values are means of ten samples. Stands were sown at 50 cm row width at marked days in 1988, but the first stand was sown already in 1987 and managed like an ordinary seed production stand in 1988.

Sowing date	Number of panicles	Dry weight (g) of panicles	Number of panicles per 15 cm*
Sowing 24.6.1987	66.4[a]	35.1[ab]	19.9
Sowing 18.5.1988	76.5[a]	36.8[a]	23.0
Sowing 1.6.1988	71.1[a]	35.6[ab]	21.3
Sowing 15.6.1988	36.6[b]	23.0[bc]	11.0
Sowing 29.6.1988	37.3[b]	19.5[c]	11.2
Sowing 13.7.1988	37.7[b]	20.1[c]	11.3
Sowing 27.7.1988	25.9[bc]	13.7[cd]	7.8
Sowing 10.8.1988	4.2[c]	4.2[d]	1.3

[a–c] Means with a different superscript letter within a column are significantly different ($P < 0.05$).

* The calculated number of panicles per 15 cm is presented to make it easier to compare the results of Tables 2, 3 and 4 with each other.

Table 5. Effect of sowing date on the number of produced panicles and tillers, and on the dry weight of panicles and above ground phytomass of cocksfoot. One seed was sown per pot on mentioned dates. Growing in glasshouse at 24 h daylength at 18 °C temperature began 24.11.1989. The trial was terminated 22.1.1990. n = 12. Values per pot.

Sowing date	Number of panicles	Dry weight (g) of		Number of large tillers at	
		panicles	phytomass	start	end
Sowing 6.6.1989	41.3[a]	13.4[ab]	63.0[a]	13.0[a]	45.4[a]
Sowing 20.6.1989	35.8[ab]	13.6[ab]	66.3a	12.7a	43.9[a]
Sowing 4.7.1989	40.4[a]	14.5[a]	71.0[a]	12.8[a]	46.8[a]
Sowing 18.7.1989	26.8[b]	9.7[b]	58.0[a]	7.0[b]	36.5[a]
Sowing 1.8.1989	11.8[c]	3.3[c]	38.0[b]	1.3[c]	23.9[b]

[a–c] Means with a different superscript letter within a column are significantly different ($P < 0.05$).

included also dead tillers. In the stands of the 1st and 2nd sowing dates a great number of lateral tillers were present in the samples. It was difficult to define the developmental stage of the plants in the fall. Every stand had tillers of different size and age. The number of leaves of the largest tillers was at least 7 in all sowing dates at the beginning of the growing period in the glasshouse. In the pot Trial II, the structure of the stands of the first three sowings was quite similar. The moderate basic fertilization of the stands was obviously not big enough to support the tillering and vegetative growth of the two oldest stands during the summer and fall.

The number of panicles, number of main
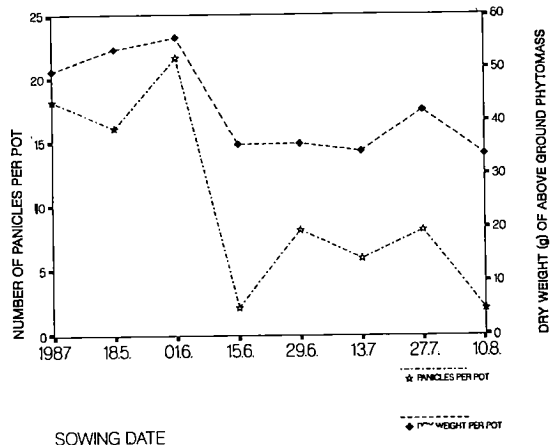
Fig. 1. The effect of sowing date in 1988 on the number of panicles and on the dry weight of above ground phytomass of cocksfoot in the subsequent spring after sowing. Sample stands were grown in a glasshouse. Values per pot.

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tillers, dry weight of panicles, and that of above ground phytomass in both trials is presented in Tables 2—5. Delaying the sowing date affected the panicle production and the above ground phytomass production of stand differently and this is shown in Fig. 1. The first two sowing times produced a significantly higher number of panicles than the last two sowing times in both trials (Tables 2—5). In the pot seeded trial floral initiation was observed to occur in the most advanced tillers already in the fall before the growing period in the glasshouse had started.

DISCUSSION

The floral evocation of cocksfoot has been detected to occur in various environmental conditions (GARDNER and LOOMIS 1953, CALDER 1963, 1964 b, HOVIN et al. 1966, WILSON and THOMAS 1971, BLONDON 1972, 1985, KOZUMPLIK and CHRISTIE 1972 b, BROUE and NICHOLLS 1973, IKEGAYA et al. 1980, HEIDE 1987). Therefore, it is difficult to determine exactly when the conditions favourable to floral evocation began, and the duration of the inductive period for the stands of each sowing date. Although the critical temperature for primary induction can be as high as 21 °C (BLONDON 1972, HEIDE 1987) the assumption, based on the climatological data, is made that the stands of all sowing times had an equally long period in optimal evocative conditions. The temperature fell into the inductive area of +10 to +2 °C (BLONDON 1985) in the middle of September when the stands of the all sowing dates had been well established. Daylength decreased into the inductive short daylength area (from 13 to 8 h) also at about the middle of September. The study on the development of the ability of cocksfoot to produce panicles in Jokioinen also indicates that floral evocation is incomplete before the end of September (NIEMELÄINEN 1990). Therefore the evocative period has been estimated to last in both falls approximately about 7 weeks (from the 15th of September to the 24th of November).

The effects of tillering and juvenile phase on the number of panicles are confounded in the present data. The number of largest tillers (diameter at base >5 mm) was counted in order to reveal the number of 'main tillers' for each sowing date. However, the high increase in the number of 'main tillers' from the start of the growing period in the glasshouse to the end of it suggests that the counting of 'main tillers' at the termination of the trial does not give accurate information of the situation at the beginning of the growing period. In spite of the problems of confounded effects, the slight differences in the number of 'main tillers' compared to the differences in the number of the panicles in different treatments indicate the presence of the juvenile phase in this trial. Stands sown on different date reacted differently to equally long evocational period and produced a different number of panicles per pot. Juvenility considerably decreased the panicle formation of stands sown in late July or early August. Experiences with the establishment of cocksfoot seed production stands in Finland (VALLE 1963, HAKKOLA 1967, ANTILA 1971, NIEMELÄINEN, unpubl.) indicate that the seed yield in the first harvest year has been considerably larger in those stands that have been established in spring without a cover crop or in an early harvested cover crop, than in stands established in a late harvested cover crop or in July without a cover crop.

In Trial II those tillers which were small (base diamater <5 mm) at the beginning of the glasshouse growing period were also able to pro-

duce panicles. According to IKEGAYA (1984) the induction stimulus of main stem of cocksfoot can be transmitted to the apicies of lateral tiller buds to some extent and this may have increased the production of panicles of small tillers. According to IKEGAYA et al. (1981 a, b) the size and the age of the cocksfoot seedling affect sensitivity to inductive conditions. The winter period increased panicle formation especially in the late sown stands and in the stand established already in 1987. This suggests that young seedlings and tillers which arose from lateral buds needed a longer time for floral evocation than larger main tillers. Probably the beneficial conditions of winter and spring for floral evocation (NIEMELÄINEN 1990) are the most important for the young seedlings and tillers. The importance of summation of different evocational factors when the level of each factor is under the threshold level has been stressed by BERNIER (1988).

The negative effect of delayed sowing was much weaker on above ground phytomass production than on panicle production. This suggests that juvenility may be the main reason for situations common in Finland when the seed production stand is dense and produces phytomass normally but does not produce panicles (KÖYLIJÄRVI 1983).

Because the whole stand in Trial II was based on the growth and tillering of one seed, it indicates that tillering has a dramatic influence on the number of panicles in the subsequent year of establishment. The small optimum number of plant density (from 50 to 60 plants m^{-2}) in the seed rate trials of NORDESTGAARD (1979) also indicated the important role of tillering. The importance of the tillers produced early in the fall to subsequent years' inflorescences in cocksfoot and in other grasses has been stressed by LANGER and LAMBERT (1963), SCHÖBERLEIN (1987), NORDESTGAARD (1988), and HEIDE (1990).

The most advanced tillers initiated flower primordia already in the fall in the pot seeded experiment. The apex was at the transition stage or just after it (stages 3 and 4) according to the scale of JEATER (1956). This observation was unexpected, since the cocksfoot material used in the experiment of HEIDE (1987) did not start floral initiation at 8 hour daylength at + 15 °C temperature during a 4-week growing period. A further trial was conducted to determine the critical day-length of cv. Haka for floral initiation. The results indicated that the critical daylength for floral initiation was about 10 hours but a long 9-week growing period at + 15 °C temperature was needed (NIEMELÄINEN 1990). HEIDE (1990) has stressed the great variation in the critical daylength and temperature conditions, and especially in the duration of evocative period between ecotypes within each species. A possible floral initiation before winter in those stands which are established early in spring without a cover crop needs further examination although floral initiation in the fall on a larger scale in the field grown material is not probable. The initiation of flower primordium in the fall causes a risk for the wintering of tillers (GARDNER and LOOMIS 1953).

We conclude that the juvenility of cocksfoot affects negatively panicle production in the subsequent year of establishment. The effect of juvenility increases as the sowing date is delayed and when the number of large tillers in the fall is decreased. Juvelinity may have an influence on the panicle production of older cocksfoot stands, too. Because of juvenility, the panicle production of the stand can be poor although the stand produces vegetative phytomass abundantly.

The juvenility characteristics of cocksfoot must be taken in to consideration when a seed production stand is established and also when the fall management of harvested seed production stand is planned.

Nowadays the establishment of a seed production stand without a cover crop is economically possible because the state pays a subsidy to farmers who keep arable land fallow for one

year (ANON. 1989). According to the regulations, it is possible to establish a seed production stand of grasses during such a fallow year (ANON. 1990 a, b). The high number of panicles produced from the early sowings in these trials suggests that such a method of establishing a

seed production stand is highly recommendable because the economic outcome is secured through the support of the state during the establishment year, and the probable seed yield is quite high in the first seed production year.

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SELOSTUS

Koiranheinän *(Dactylis glomerata* L.) röyhynmuodostukseen vaikuttavista tekijöistä Suomessa

II. Nuoruusvaiheen, kylvöajan ja versonnan vaikutus.

OIVA NIEMELÄINEN

Maatalouden tutkimuskeskus

Nuoruusvaiheen merkitystä kylvövuoden jälkeisen vuoden röyhynmuodostukselle tutkittiin kylvämällä koiranheinä-kasvustoja kasvukauden eri aikoina. Ensimmäisessä kokees-sa Haka koiranheinä kylvettiin pellolle Jokioisissa kahden viikon välein 18.5.1988 ja 10.8.1988 välisenä aikana. Kas-vustojen röyhynmuodostuskyky tutkittiin näytekasveja kas-vihuoneessa kasvattamalla sekä ennen talvea että talven jäl-keen. Lisäksi määritettiin pellossa kasvaneiden kasvustojen röyhynmuodostus. 18.5.1988 ja 1.6.1988 kylvetyt kasvustot tuottivat merkitsevästi enemmän röyhyjä kuin myöhemmin kylvetyt kasvustot.

Toisessa kokeessa suoraan astioihin alkukesällä kylvetystä yhdestä siemenestä muodostuneet kasvustot versoivat kyl-vövuonna voimakkaasti, ja tuottivat kylvövuoden syksyl-lä kasvihuonekasvatuksessa yli 40 röyhyä astiaa kohden. Astioihin kylvettyjen kasvustojen röyhynmuodostus oli run-sasta vielä kylvöaikana 18.7.1989 ja 1.8.1989 perustetuis-sa kasvustoissa.

Kylvöajankohdan myöhästyttäminen vähensi voimak-kaammin koiranheinän röyhynmuodostusta kuin maanpääl-lisen kasvimassan tuotantoa. Koiranheinän nuoruusvaiheen olemassaolo ja siitä johtuva vaillinainen kukinnan viritty-minen voi vähentää röyhynmuodostusta erityisesti kylvö-vuoden jälkeisenä vuonna, mutta nuoruusvaiheella saattaa olla merkitystä myös myöhäisempien tuotantovuosien röy-hynmuodostukselle. Nuoruusvaiheen vaikutus on yksi to-dennäköinen tekijä niissä tilanteissa, joissa koiranheinän sie-menviljelykasvusto tuottaa vihermassaa runsaasti mutta röy-hyjä vain vähän.

Yhdestä siemenestä muodostuneet kasvustot tuottivat runsaasti röyhyjä ja riittävän suurella ja oikea-aikaisella ver-sonnalla on huomattava merkitys koiranheinän röyhynmuo-dostukselle.

Koiranheinän nuoruusvaiheen olemassaolo ja versonnan vaikutus röyhynmuodostuksessa tulee ottaa huomioon pe-rustamistapaa valittaessa. Satovuotta edeltävänä syksynä kas-vustossa tulisi olla runsaasti riittävän suuria versoja runsaan röyhynmuodostuksen aikaansaamiseksi. Siemennurmen pe-rustaminen viherkesantona on yksi suositeltava perustamis-tapa. Kasvusto on kylvettävä riittävän aikaisin keväällä.

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FACTORS AFFECTING PANICLE PRODUCTION OF COCKSFOOT (DACTYLIS GLOMERATA L.) IN FINLAND.

III. RESPONSE TO EXHAUSTION OF RESERVE CARBOHYDRATES AND TO FREEZING STRESS

OIVA NIEMELÄINEN

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NIEMELÄINEN, O. 1990. Factors affecting panicle production of cocksfoot _(Dactylis glomerata_ L.) in Finland. III. Response to exhaustion of reserve carbohydrates and to freezing stress. Ann. Agric. Fenn. 29: 241—250. (Agric. Res. Centre, Inst. Crop and Soil Sci., 31600 Jokioinen, Finland.)

Plants of cocksfoot cv. Haka were taken just prior to the onset of winter and subjected to cold treatment at −20 °C temperature in laboratory. Panicle production and growth of the plants was analyzed after an 8-week growing period.

Panicle number and dry weight of above ground phytomass was significantly smaller when plants were without a snow cover than when plants were under a 20 thick snow cover or under an ice cover. The negative effect was stronger on the number of panicles than on the above ground dry weight of phytomass. Growth and panicle production of plants which were under snow or ice cover did not differ significantly from the growth of control plants which were kept at + 3 °C temperature during the cold treatment.

In the other study the stress caused by a lengthly snow cover period was simulated by keeping flower-induced plants various time in the dark at + 10 °C. This treatment caused exhaustion of plants reserve carbohydrates. In spring sampled plants a 28-day stress period decreased panicle production significantly compared to plants without stress. However, plants sampled in the fall did not respond negatively even to a 42-day stress period. The negative effect of the exhaustion of the plant's reserve carbohydrates was stronger on the number of panicles produced than on the above ground dry weight of phytomass.

Both cold temperature during winter and excessively long snow cover are a greater risk to the panicle production ability in cocksfoot than to phytomass production. Proper winter hardening in the fall, and good condition of stand in the spring is particularly important to the cocksfoot seed production stand.

Index words: _Dactylis glomerata,_ orchard grass, seed production, freezing stress, cold tolerance, stress, floral evocation.

INTRODUCTION

Good wintering ability is a prerequisite for satisfactory performance of perennial grasses in Northern conditions. In this study the effect of winter was examined from the point of view of how conditions during winter might have a specific negative effect on the panicle production of cocksfoot. Two factors have been studied. Firstly, the effect of low temperature,

and secondly the effect of exhaustion of the plant's reserve carbohydrates.

Low temperature is one factor which can damage the growing point of grasses so that panicle formation is inhibited. In the trial of GARDNER and LOOMIS (1953) many cocksfoot plants were injured by freezing in the fall after induction requirements had been fullfilled. A typical reaction was a return to vegetative growth. Growth continued from axillary buds but no panicles were formed on the new tillers. Similar results have been reported concerning red fescue and Kentucky bluegrass (HODGSON 1966), and bromegrass (SASS and SKOGMAN 1951, ELLIOT 1966). Usually the initiated inflorescences have been destroyed but the vegetative growing points, on the contrary, have survived as in HEIDE's (1980) laboratory experiments with Kentucky bluegrass.

In addition to cold temperature, plants are subjected to such stress during a lengthly winter that a considerable amount of a plant's reserve carbohydrates (RCH) is consumed. The number of panicles per plant is usually higher when the plants remain for a longer period in inductive or vernalisive conditions (HÅBJØRG 1978, 1979). However, HÅBJØRG (1979) has ob-served that too long a time in an inductive short day led to a decrease in the number of panicles in Kentucky bluegrass. According to HÅBJØRG the decrease was caused by the exhaustion of RCH during warm weather in the fall when the plants were in winter dormancy. GARDNER and LOOMIS (1953) studied the effect of the exhaustion of the carbohydrate reserves of cocksfoot on panicle production by storing induced plants in darkness until the plants lost their green colour. When the plants were transferred to continuous light, they resumed their green colour rapidly and started to grow but formed no panicles. The plants lost their ability to form panicles possibly due to a lack of reserve carbohydrates.

The aim of this study was to clarify if low temperature or exhaustion of a plant's reserve carbohydrates during winter can inhibit the panicle production of cocksfoot without simultaneously harming vegetative growth. The target was to elucidate the reasons for the phenomenon observed in Finland (KÖYLIJÄRVI 1983) in which cocksfoot seed production stands produce vegetative phytomass normally but do not produce panicles.

MATERIAL AND METHODS

Two different series of trials were conducted. In all trials field grown and naturally induced sample plants were used. Row seeded Haka cv. stand was grown at the Institute of Crop and Soil Science of the Agricultural Research Centre in Jokioinen. Fifteen cm sections of a row were dug up as the sample stand. The samples were then planted in pots of soil.

In the cold temperature treatments samples were taken just before the soil froze in the fall. (16.11.1988 in Trial AI and 24.11.1989 in Trial AII). The content of reserve carbohydrates was analyzed when the sample plants were taken. The low temperature treatments were performed in freeze rooms and the plants were subsequently grown in a glasshouse.

The samples were transferred into the freeze room to 0 °C temperature. In the freeze room the pots were placed into a container and the temperature at the bottom of the container was adjusted to 0 °C for the whole treatment period. The height of the pots was 19.5 cm. The aim of the procedure was to avoid excessive low soil temperatures thereby preventing injury

to the roots and to describe more accurately natural soil temperatures. The pots were enveloped with peat soil.

The following treatments were performed. Control pots (A) were kept at $+3$ °C during the whole treatment. In treatment B the samples were kept as such in bare ground. In treatment C a 20 cm thick snow cover was formed on the samples at the beginning of the low temperature treatment. In treatment D a 5 cm thick ice cover layer was frozen on the sample plants before the low temperature treatment began. In Trial AII only treatments A, B and C were conducted. 10 pots were used per treatment. Treatments B, C and D remained in the same freeze room throughout the whole treatment period.

The temperature in the freeze room was lowered gradually so that the minimum temperature -20 °C lasted for 72 hours. Duration of the treatment was 7 days. After the treatment, the temperature was increased to $+3$ °C for one day and then the plants were transferred into the glasshouse where they were kept for 7 days at $+5$ °C and in natural light conditions. After this,. the plants were fertilized with a mixed fertilizer (17-6-12 NPK) at 5 grams per pot and grown at about $+18$ °C in continuous light (80 Wm^{-2} PAR + natural light). The location of the pots in the glasshouse was according to the randomized block design. The trials were terminated after 8 weeks growing period in the glasshouse. Most of the plants were at the flowering phase by the termination of the trial.

Three trials were conducted concerning the exhaustion of plant reserve carbohydrates. The main difference between the trials was that the sample plants were taken just after winter in Trials BI (28.4.1988) and BIII (20.4.1989) but just prior to the onset of winter (16.11.1988) in Trial BII. The exhaustion of reserve carbohydrates was achieved in such a way that the sample plants were kept for different times in darkness at $+10$ °C temperature when taken from the field. The control plants (A) were moved directly to growing conditions in a

growth cabinet in all experiments. Darkness stress lasted from 14 days to 49 days in the different treatments and trials (see Tables 3—5). The possible devernalizing effect of the $+10$ °C temperature was examined by keeping the samples at 10-hour daylength at $+10$ °C temperature for 28 days in Treatment E of Trial BII. After the stress period, the growing period lasted 33 days in continuous light at $+18$ °C temperature. The pots were fertilized with a 5 g mixed fertilizer 17-6-12 NPK at the beginning of cultivation in the growth cabinet. Irradiance was 105 Wm^{-2} (PAR) at plant level. The plants were located in separate growth cabinets because otherwise the plants removed later would have been shaded by those removed earlier.

After the stress treatments had begun, the fungicide Bayleton 25 (a.i. triadimefoni 250 g kg^{-1}) was sprayed at 0.03 % solution on the plants to prevent the invasion of fungal diseases.

The RCH content of plants was analyzed at the beginning of the growing period in Trials BII and BIII. A 3-cm-long piece from the base of the main stem was taken from additional sample plants and the content of RCH analyzed according to the following method of PULLI developed at the Institute of Crop and Soil Science by modifying the methods of NELSON (1944) and SOMOGYI (1952). The method measures glucose, fructose, saccharose and fructosans.

In the reserve carbohydrate analyses 0.5 grams of dried sample was weighed in erlenmyer bottles. The bottles were filled to 100 ml with 0.1 N HCl and inverted overnight at $+50$ °C. The solution was filtered into 250 ml bottles. The sediment was washed with water and the bottle filled to 250 ml. 40 ml of this filtrate was measured on ion-exchangers (4 g + 4 g) in a 100 ml erlenmyer. The mixture was then shaken for 1 hour and the ion-exchangers filtered away. The filtrate was diluted 1:10. Two ml of the diluted solution was measured in test tubes. Two ml of copper-reagent was added

and the test tubes then filled to 7 ml with distilled water. The tubes were mixed and boiled for 20 minutes. After cooling, 2 ml of arsenomolybdate was added. The test tubes were filled to 10 ml with water. The amount of reserve carbohydrates was measured with a spectrophotometer (Shimadzu) at the 540 nm wavelength. Contents were determined using a standard curve.

In both series of trials the number of panicles, the dry weight of panicles and that of the above ground phytomass were measured per pot at the termination of the trial. The number of large tillers (diameter >5 mm at the base of stem) was counted. The significance of the differences of means was tested using Tukey's multiple comparison test (SNEDECOR and COCHRAN 1978).

RESULTS

The cold temperature treatment at $-20\ °C$ significantly decreased panicle production, above ground phytomass production, and also the number of large tillers when the plants were kept on bare ground in comparison to the control plants and the plants covered with snow (Tables 1 and 2). The panicle production of control plants and the snow covered plants did not differ significantly. In this material a stronger negative effect was found for panicle formation (68 % in Trial AI and 90 % in Trial AII) than for phytomass production (41 % in trial AI and 58 % in trial AII), but significant and strong also in phytomass production. Freezing injury was not confined to panicle production only but also had a negative effect on phy-

Table 1. The growth and panicle production of cocksfoot after 7-day cold treatment at $-20\ °C$ and subsequent 8-week growing period in continuous light at $+18\ °C$. n = 10. Trial AI. Values per pot.

Treatment*	Number of panicles	Dry weight (g) of		Number of tillers
		panicles	phytomass	
A. Control	14.5[a]	3.5[a]	59.1[a]	27.5[a]
B. Bare ground	4.7[b]	1.1[b]	34.7[b]	15.3[b]
C. 20 cm snow cover	20.2[a]	4.0[a]	56.7[a]	30.9[a]
D. 5 cm ice layer	14.7[a]	3.2[a]	57.9[a]	28.2[a]

[a–b] Means with a different superscript letter within a column are significantly different (P<0.05).

* Control plants were kept at $+3\ °C$ during the treatment. In treatment B the plants were on bare ground, in treatment C under a 20 cm thick snow layer, and in treatment D a 5 cm thick ice layer was formed on the plants before cold treatment.

Table 2. The growth and panicle production of cocksfoot after 7-day cold treatment at $-20\ °C$ and subsequent 8-week growing period in continuous light at $+18\ °C$. n = 10. Trial AII. Values per pot.

Treatment*	Number of panicles	Dry weight (g) of		Number of tillers
		panicles	phytomass	
A. Control	23.5[a]	4.6[a]	57.8[a]	31.3[a]
B. Bare ground	2.4[b]	0.5[b]	24.0[b]	11.1[b]
C. 20 cm snow cover	24.8[a]	4.8[a]	60.8[a]	33.5[a]

[a–b] See Table 1.

* Control plants were kept at $+3\ °C$ during the treatment. In treatment B the plants were on bare ground and in treatment C a 20 cm thick snow layer was formed on the plants before cold treatment.

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tomass production and tiller number.

In the study on exhaustion of plant RCH, leaf elongation started in darkness at +10 °C temperature. Gradually the colour of the leaves turned yellow. Growth began earlier and the visually evaluated condition of plants decreased more rapidly in plants sampled in the spring than in the samples taken in the fall. In the spring samples, the 49-day stressed plants lost their green colour completely in Trial BI and the plants died before the 42-day stress period was over in Trial BIII.

The content of RCH before the start of growing period in the growth cabinet for different treatments in Trials BII (fall samples) and BIII (spring samples) was:

	Content of reserve carbo-hydrates % in dry matter	
	Trial BII	Trial BIII
A. Stress 0 days	52.1	13.0
B. Stress 14 days	44.0	10.5
C. Stress 28 days	34.3	7.6
D. Stress 42 days	30.1	plants died
E. 28 days at 10 h daylength	48.0	

The effect of darkness stress on panicle number was much stronger for spring samples than fall samples (Tables 3—5). In Trial BI stress period of 29 and 49 days and in Trial BIII a 28-day stress period decreased panicle number signifi-

Table 3. The growth and panicle production of cocksfoot after a stress period in darkness at +10 °C and subsequent 33-day growing period in continuous light at +18 °C. The sample plants were taken from the field on 28.4.1988. n = 12. Trial BI. Values per pot.

Treatment	Number of panicles	Dry weight of above ground phytomass (g)	Number of main tillers
A. No stress	24.5[a]	48.0[a]	30.3[a]
B. Stress 29 days	5.3[b]	40.6[a]	21.1[b]
C. Stress 49 days	0.2[c]	17.8[b]	6.5[c]

[a–c] See Table 1.

Table 4. The growth and panicle production of cocksfoot after a stress period in darkness at +10 °C and subsequent 33-day growing period in continuous light at +18 °C. The sample plants were taken from the field on 16.11.1988. n = 12. Trial BII. Values per pot.

Treatment	Number of panicles	Dry weight of panicles (g)	Dry weight of above ground phytomass (g)	Number of main tillers
A. No stress	7.6[bc]	1.7[ab]	66.2[a]	27.5[a]
B. Stress 14 days	11.0[ab]	1.9[ab]	63.4[a]	25.1[a]
C. Stress 28 days	13.2[ab]	2.7[a]	79.5[a]	23.8[a]
D. Stress 42 days	3.6[c]	0.9[b]	72.2[a]	26.5[a]
E. 28 days at 10 hour daylength at +10 °C	18.0[a]	3.2[a]	77.1[a]	31.8[a]

[a–c] See Table 1.

Table 5. The growth and panicle production of cocksfoot after a stress period in darkness at +10 °C and subsequent 33-day growing period in continuous light at +18 °C. The sample plants were taken from the field on 20.4.1989. n = 12. Trial BIII. Values per pot.

	Number of panicles	Dry weight of panicles (g)	Dry weight of above ground phytomass (g)	Number of main tillers
A. No stress	11.1[a]	1.7[ab]	27.3[b]	17.3[a]
B. Stress 14 days	7.3[a]	2.1[a]	66.4[a]	22.5[a]
C. Stress 28 days	1.5[b]	0.5[b]	38.2[b]	18.9[a]

[a–c] See Table 1.

cantly. However, in Trial BII with fall samples not even the 42-day stress period decreased the number of panicles significantly. The stress treatments had a significant effect on the weight of above ground phytomass only in Trial BI with a 49-day treatment. The number of main tillers decreased significantly, too. The reason for the substantial differences in the dry weight of above ground phytomass in Trial BIII is unclear.

Short daylength prevented the loss of RCH during the treatment and the number of panicles produced per pot was high (treatment E in Trial II, Table 4).

DISCUSSION

Weather conditions were good for hardening during both fall seasons for at least one month before the plants were sampled. According to KACPERSKA-PALACS (1978) temperatures from 5 °C to 2 °C are good for the first phase of hardening and the temperatures from 0 °C to −2 °C are beneficial to the second phase of hardening. The hardening process is most active during the first week of hardening (GUSTA 1986). The content of reserve carbohydrates in the sample plants was high (52 % of d.m. in Trial IA and 43 % in Trial IB) indicating that the plants were in a well-hardened state when the cold temperature treatments were conducted.

The data indicate that −20 °C temperature decreases both the panicle and the phytomass production of hardened cocksfoot plants when the stand lacks a protective snow cover. The protective effect of snow cover (JAMALAINEN 1970, ANDREWS 1987) was evident in this experiment. The cold hardiness of cocksfoot is poor (GUDLEIFSSON et al. 1986, LIMIN and FOWLER 1987), and the lack of a snow cover was very detrimental to the wintering of cocksfoot in YLIMÄKI's (1962) trial in southern Finland. When the snow cover was artificially removed, the wintering of cocksfoot decreased from 7.3 to 0.1 (on the scale from 10 to 0).

According to STOUT et al. (1988) the most direct way to obtain an estimate of frost injury is to dig plants from the field, plant them in pots of soil, and then expose the plants to optimum growing conditions in a glasshouse. We can compare the results of laboratory experiments, to some extent, with the observations of the row seeded cocksfoot stand in natural conditions. The row stands from which the samples were taken survived well in the natural conditions when we take into consideration the low temperatures that occurred in the fall of 1988 (see NIEMELÄINEN 1990 for weather data). Those sample plants which were taken in the spring of 1989 from the same stand produced as many panicles per pot (12.2—18.2) as the control plants sampled in the fall in this trial (14.5) (NIEMELÄINEN 1990). In natural conditions, just a thin layer of snow and the upward flux of soil warmth can strongly affect temperature conditions at the soil surface. The results support the statement of LARSEN (1986) that inspite of the advantage of laboratory tests where material can be tested in a relatively short time, laboratory tests are simple in comparison with the complex situation in nature.

The results support the findings of LÜTKE-ENTRUP and SCRIMPF (1964) that the occurrance of low temperatures when the soil lacks a snow cover decreases the seed yield of overwintering grasses. HEIDE (1980) suggested that a steady snow cover protects the inflorescences against freezing. Freezing has been recognized to be a problem in areas where snow cover is only occasional. For example in Britain it is recommended to leave some regrowth before winter to protect the growing points of cocksfoot

against extreme cold (ANON. 1978).

It is not possible to evaluate the tolerance of cocksfoot to prolonged ice cover by the results of this trial. The duration of the ice cover treatment was so short that such a stress factor as unaerobic conditions did not have time to develop. The ice cover did not increase susceptibility to short-term low temperature. The ice tolerance of cocksfoot is low in general (TEITTINEN 1958, GUDLEIFSSON et al. 1986, PULLI 1987).

The ability of grasses to withstand low temperature varies during winter (PULLI 1986). Frost hardiness is best during early winter but decreases rapidly in the spring (GUSTA 1986). In the present cold temperature experiments the sample material was taken in the fall. In the spring, already temperatures milder than -20 °C may have a negative effect both on the panicle and the phytomass production of cocksfoot.

The exhaustion of reserve carbohydrates was performed by storing the plants in darkness at moderate temperature. Such a procedure has been used for testing the vitality of plants (HUOKUNA 1964), and the amount of etiolated growth has been used for the estimation of plant food reserves (KLEBESADEL 1985, McKENZIE et al. 1988). The natural winter was not used in the present study because many different factors act simultaneously during winter, and the effect of the exhaustion of reserve carbohydrates is difficult to separate from other negative or positive factors.

The content of RCH in the unstressed plants sampled in the fall was 52.1 % of dry matter. The level of RCH decreased in the stress treatments but the decrease was slight compared to the effect of natural winter. The RCH content of fall samples stressed for 42 days was 30.1 % but the RCH content of overwintered plants (without an extra stress period) was only 13.0 %. RCH content was measured in the spring taken samples (20.4.1989) just prior to the start of spring growth, but it is not out of the question that some spring growth processed had already begun and decreased the RCH further. RCH content is at a minimum just when spring growth begins (WALTON 1983, p. 146). The dramatic decrease of RCH during winter has been observed in other perennial grass species, too, in Jokioinen (PULLI unpubl.). HUOKUNA (1964) reported values for RCH content of stem bases of cocksfoot from 37 to 64 % in the fall and from 17 to 27 % in July, which are quite near to the values in this study.

Plants sampled in the fall survived the stress treatments better than the spring samples. Apparently the lower level of RCH before the stress treatment and the rapid start of growth in the darkness resulted in a rapid decline in the condition of the spring samples compared to the fall samples.

In the fall samples the transfer of control plants from slightly frozen soil directly to $+18$ °C may have disturbed the growth of the plants, and decreased the number of panicles in the control treatment to some extent. On the other hand, the short stress period at $+10$ °C may have also increased the floral evocation of the fall taken sample plants. Floral evocation has been incomplete before the onset of winter in Jokioinen (NIEMELÄINEN 1990) and the $+10$ °C temperature has been inductive in short daylength conditions (IKEGAYA et al. 1979 and 1982, HEIDE 1987).

The possible devernalizing effect of high temperature (BERNIER et al. 1981) was checked by the treatment in short daylength at the same temperature at which the stress treatments were conducted. Short daylength prevented RCH loss, and the abundant number of panicles indicate that the $+10$ °C was not so high a temperature that it could have destroyed the vernalized or induced condition.

The results support the findings of GARDNER and LOOMIS (1953) that exhaustion of a plant's RCH can lead to the loss of it's panicle formation ability although vegetative growth can be regained.

The effect of long winter in the panicle production of cocksfoot can be two sided. On the one hand, winter can strengthen the floral evocation (NIEMELÄINEN 1990). On the other hand, low temperature and a long snow cover period can stress the plant to such an extent that it does not produce panicles. Although one must be cautious in applying the findings of these pot trials to natural conditions, the data indicate that a stress caused by a long lasting snow cover or low temperature is a greater risk for panicle production than for the wintering of cocksfoot.

Freezing stress and ice sheet injuries are considerable risks to the success of seed production of cocksfoot in the traditional seed production area in southwest Finland (KÖYLIJÄRVI 1983).

In those areas it often occurs that temperatures are low when the snow cover is absent. According to fodder production trials (MUSTONEN et al. 1990) the overwintering and fodder production of cocksfoot has succeeded well in the Central areas of Finland where the snow cover is usually permanent throughout the winter. The possibilities for cocksfoot seed production in such areas of Finland where the risks of low temperature and excessively long snow cover period are small (RANTANEN and SOLANTIE 1987) should be studied in practice.

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SELOSTUS

Koiranheinän (*Dactylis glomerata* L.) röyhynmuodostukseen vaikuttavista tekijöistä Suomessa

III. Vararavintohiilihydraattien kulumisen ja pakkasen vaikutus

OIVA NIEMELÄINEN

Maatalouden tutkimuskeskus

Talven aikana esiintyvien stressitekijöiden vaikutusta Hakakoiranheinän röyhynmuodostukseen ja kasvuun tutkittiin laboratoriokokeissa. Kasveja rasitettiin −20 °C asteen pakkaskäsittelyllä tai pitkäkestoisen lumipeiteajan rasitusta kuvattiin kuluttamalla kasvien vararavintohiilihydraatteja pitämällä kasveja pimeässä + 10 °C asteen lämpötilassa. Tutkimuksessa pyrittiin selvittämään, onko talven stressitekijöillä vaikutusta siihen, että vaikka koiranheinän siemenviljelykasvusto tuottaa vegetatiivista kasvimassaa runsaasti niin röyhyjä muodostuu vain vähän.

Pakkanen (−20 °C) vähensi merkitsevästi paljaana olleiden koiranheinäkasvustojen sekä röyhynmuodostusta että maanpäällisten osien kuiva-aineen tuotantoa kontrollikasvustoihin verrattuna. Sen sijaan 20 cm:n paksuisen lumikerroksen alla olleet kasvit säilyivät käsittelyssä vahingoittumattomina. Viiden cm:n paksuisen jääkerroksen alla olleet kasvit eivät vahingoittuneet lyhytkestoisessa (7 vrk) pakkaskäsittelyssä.

Maan lämpö ja jo ohutkin lumikerros suojaavat luonnonoloissa talvehtivien koiranheinien kasvupisteitä pakkasta vastaan. Pakkanen aiheuttaa kuitenkin huomattavan riskin sekä koiranheinän röyhynmuodostukselle että talvehtimiselle. Pakkasen aiheuttama vaurio ei kohdistunut ainoastaan röyhynmuodostukseen vaikka vaikutus olikin suhteellisesti suurempi röyhynmuodostuksessa kuin koko maanpäällisen kasvimassan tuotannossa.

Tutkimuksen toisessa osassa tutkittiin kasvin vararavintohiilihydraattien kulumisen vaikutusta koiranheinän kasvuun ja röyhynmuodostukseen. Kuluttaminen tehtiin pitämällä kasveja pimeässä + 10 asteen lämpötilassa. Kasvin vararavintohiilihydraatit vähenivät stressikäsittelyn aikana, mutta eivät niin voimakkaasti kuin luonnollisen talven

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aiheuttaman rasituksen aikana. Neljän viikon rasituskäsittely vähensi keväällä otettujen näytekasvien röyhynmuodostusta merkitsevästi. Vaikutus maanpäällisen kasvimassan tuotantoon oli vähäisempi kuin vaikutus röyhynmuodostukseen.

Sekä alhaisen lämpötilan että pitkäkestoisen talven aikaansaama stressi aiheuttaa suuremman riskin koiranheinän röyhyn- kuin vihermassan tuotannolle. Koiranheinän kunnollinen karaistuminen syksyllä ja hyvä kasvukunto keväällä ovat erityisen tärkeitä siementuotantokasvustoille. Koiranheinän siementuotannon onnistumista Suomessa tulisi tutkia alueilla, missä sekä vähälumisuuden ja että liiallisen lumisuuden ja pitkän talven aiheuttama riski on pieni.

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