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N_2O, CO_2 and O_2 concentrations in soil air influenced by organic and inorganic fertilizers and soil compaction

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The soil air composition under ley was studied during the fifth year of a field experiment carried out in a humid climate and with different fertilization and soil compaction treatments. The fertilization treatments were: NPK fertilizer (220 kg NH_4NO_3-N ha^{-1}); cattle slurry (CS) (189 kg total N ha^{-1}); CS (131 kg total N ha^{-1}); CS (84 kg total N ha^{-1}); and an unfertilized treatment. The soil was experimentally compacted by two passes with a tractor, wheel by wheel, shortly before fertilization. Soil air at a depth of 7-12 cm was sampled through stationary soil air samplers. NPK fertilization resulted in consistently higher N_2O concentrations than in the other treatments. Cattle slurry applications did not significantly affect the N_2O concentrations. The soil compaction resulted in higher N_2O concentrations especially for NPK-fertilized treatments (max concentration 563 ppm). Compacted soil had lower O_2 concentrations and higher CO_2 concentrations than the uncompacted soil. There was no effect of fertilization on the concentrations of O_2 and CO_2 in soil air.

Key words: Cattle manure, denitrification, NH_4NO_3, N_2O, soil atmosphere, soil compaction

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N_2O is a greenhouse gas, and contributes to about 6% of the total global warming (Denmead 1991). The atmospheric concentration of N_2O (315 vpb) is increasing by 0.25% per year (Denmead 1991). In the stratosphere, N_2O participates in reactions leading to the destruction of atmospheric ozone (Crutzen & Ehalt 1977).

The estimated total global production is about 8 Tg N_2O-N $year^{-1}$ (Davidson 1991) of which 0.2 -2.1 Tg N_2O comes from fertilized, agricultural land (Eichner 1990). The emission of N_2O can be perceived as a leakage of intermediate products in nitrification and denitrification (Bremner & Blackmer 1978). These reactions are influenced by available organic material, nitrate supply, oxygen availability, water content, soil pH, soil temperatures and the presence of plants (Byrnes 1990, Rheinbaben 1990). Denitrification is likely to be the main source of N_2O in wet soils (Davidson 1991). Denitrification loss of

soil N is normally less than 10% (Rheinbaben 1990), but can be so great that it seriously affects the nitrogen balance in an agricultural production system. Rheinbaben (1990) refers to denitrification estimates by the ^{15}N balance and acetylene inhibition methods ranging from 0 to 77% of total fertilized N. Soil compaction stimulates denitrification through impeded oxygen diffusion (Bakken *et al.* 1987).

The aim of this investigation was to study the composition of soil air under ley crop in a humid climate, as affected by fertilization and soil compaction by tractor traffic. Investigations were carried out in a sandy loam field fertilized with three levels of cattle slurry, NPK fertilizer or unfertilized.

MATERIALS AND METHODS

The soil air composition was studied during the fifth year (growth season 1989) of a field experiment involving different fertilization and soil compaction treatments. The field experiment was located in Surnadal, Norway 25 m a.s.l., 63°00'00" N, 8°88'44" E. Soil temperature and precipitation during the period of measurement are recorded in Fig. 1.

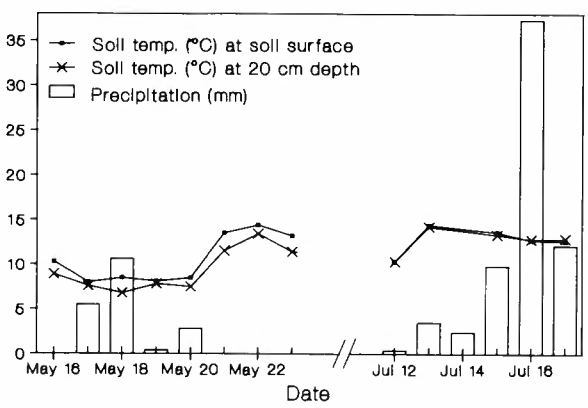

Fig. 1. Soil temperature (°C) at soil surface and at 20 cm depth and precipitation (mm) in the experimental period

The experiment was started in 1985, and was run with a crop rotation adapted to ecological farming with milk production, i.e. rich in ley and legumes. The experiment was of a split-plot factorial design with two replicates, soil compaction on main plots and fertilization on small plots (2.8 m x 8 m, sample area 1.5 m x 6.5 m). Soil air was sampled using two stationary soil air samplers (equilibrium chambers) in each plot. Thus, for each treatment, there were four parallel soil air measurements taken on each day of measurement. Fertilization and soil compaction treatments, soil, crop rotation and experimental plan are described in detail by Hansen (1992a).

The 1989 crop was a three-year-old ley with timothy and clover (*Phleum pratense L.*,

Trifolium pratense L., T.repens L., T.hybridum L.). The ley was cut twice (22 June & 23 August) and fertilizers were added on 3 May & 1 July. The fertilization treatments comprised: (**NPK**), NPK fertilizer (18% N, 3% P, 15% K) with 126 + 94 kg NH_4NO_3-N ha-1; (**CS3**), cattle slurry equivalent to 117 kg N (80 kg NH_4-N) + 72 kg N (36 kg NH_4-N) ha^{-1}; (**CS2**), 78 kg N (53 kg NH_4-N) + 53 kg N (27 kg NH_4-N) ha^{-1}; (**CS1**), 43 kg N (29 kg NH_4-N) + 41 kg N (21 kg NH_4-N) ha^{-1}, and (**Unf**), an unfertilized treatment. The cattle slurry was diluted with water up to 200% of the original volume and spread by can with a small spreading plate. The slurry was mixed continuously. NPK fertilizer was spread by hand. Samples of cattle slurry were frozen for later analyses following the method described by Horwitz (1980).

The diluted slurry contained 42 g dry matter l^{-1}, 2.1 g Kjeldahl-N l^{-1}, 1.2 g NH_4-N l^{-1}, 0.03 g NO_3-N l^{-1}, 1.8 g ether soluble substances l^{-1}, 10.5 g crude fibres l^{-1} and had a pH of 7.1.

Soil compaction treatment comprised one pass of a three tonne tractor total weight (2.5 tonne on rear wheels), wheel by wheel, once each spring, and two passes shortly after each harvest. Rear wheel inflation pressure was 150 kPa and tyre width 32 cm.

Soil samples (0-20 cm depth) were taken for mineral-nitrogen when the plant had reached a height of about 20 cm (20 May) and shortly after each harvest (23 June and 22 August). The samples were air dried at 30°C and stored for four to six months. NH_4-N and NO_3-N were extracted with 1 N KCl (10 g soil to 50 ml 1N KCl). NO_3-N was analysed by the hydrazine reduction method (Kempers & Luft 1988) and NH_4-N by the modified Berthelot reaction (Krom 1980).

Air-filled porosity was determined in undisturbed soil samples taken by means of four 100 cm^3 cylinders in each plot at a depth of 7-11 cm shortly after soil compaction (30 August). Particle density, used to calculate material and pore volume, was determined following the method of De Boodt *et al.* (1967).

The earthworm biomass was determined at a depth of 0-20 cm on 28 June (Hansen 1992a).

Gas measurements

Equilibrium chambers for soil air sampling at a depth of 7-12 cm were installed shortly after each fertilization. Samples were taken from the equilibrium chambers on seven occasions during the growth season (16, 19, 22 May and 12, 14, 15, 17 July). The chambers were constructed of pointed cylinders of brass 19.5 cm in length, and with a 6 mm outer and a 4 mm inner diameter, and eleven 1 mm holes. Gas samples were taken through a rubber septum at the top of the chambers with a two-way blood collection needle. The needle was pierced through the rubber septum and connected to a 3.4 ml evacuated vial (Venoject tube, Terumo Europe N.V., 88A11LA EXP 12 90) as outlined by Magnusson (1989). The vials were covered with a layer of silicone. The samples were stored frozen (-18°C) for one year before being analysed.

The gas samples were analysed on a gas chromatograph (Fractovap 4200, Carlo Erba, Italy) equipped with a Poraplot Q widebore 0.53 mm x 25 m column (Chrompac, Middelburg, The Netherlands). The temperature in the oven was at 31°C. Oxygen and N_2O were analysed by Electron Capture Detector (Carlo Erba Model ECD 400) with a detector temperature of 350°C. CO_2 was detected by Thermal Conductivity Detector (Carlo Erba

model HWD 450). The carrier gas (He) flow was 7 ml min^{-1}. The ECD signal for O_2 was calibrated with standards of known O_2 concentrations (10-20% O_2). The N_2O response was calibrated with purchased standards ranging from 1 to 160 vpm N_2O.

Experimental plan, statistical analyses and calculations

The main effects and interactions between soil compaction, fertilization and date were tested with analyses of variance (ANOVA) and the Newman-Keul's test. The interaction replicate*compaction was used as an error term to test effect of compaction. The correlations between any of the parameters were examined using the Corr procedure (SAS Institute 1988). Mean values for N_2O, CO_2 and O_2 for each field plot were used to investigate the correlation between these variables and earthworm biomass and air-filled porosity.

Studentized residuals were used to test the normality of the distributions with residual plots and procedure Univariate (SAS Institute 1988). None of the N_2O, O_2 or CO_2 concentrations in soil air were normally distributed, but O_2 and CO_2 concentrations were so close to normal distribution that they were treated statistically as if that were the case. Soil air concentrations of N_2O were close to lognormal distribution, and data transformed with natural logarithms were used in both variance and correlation analyses.

The means and standard errors of the means of N_2O data were calculated according to Finney's method, described by Parkin *et al.*(1988).

RESULTS

The highest N_2O concentrations in soil air were found in NPK-fertilized treatments ($p < 0.01$, Fig. 2). This effect was found in both uncompacted and compacted treatments, but the highest concentrations were found in compacted treatments. The average N_2O concentration in compacted NPK-fertilized plots was about 100 times higher than those in unfertilized plots or plots fertilized with cattle slurry. The applications of cattle slurry did not significantly affect the N_2O concentrations (Newman-Keul's test, $\alpha = 0.05$).

The N_2O concentration in soil air varied throughout the growing season (Fig. 2) with a statistically significant interaction between the fertilizer type, soil compaction and time ($p < 0.01$). In the spring, N_2O concentrations peaked later in the cattle slurry-fertilized compacted treatment than in the other treatments.

The concentrations of O_2 and CO_2 (Fig. 3) in the soil were negatively correlated ($r = -0.74$, $p < 0.01$). The soil compaction increased the CO_2 concentration ($p =< 0.01$) and lowered the O_2 concentration ($p = 0.1$) compared with uncompacted soil (Fig. 3). There was no significant effect of fertilization on the concentrations of O_2 and CO_2. The soil compaction resulted in a lower number of earthworms and reduced porosity of the soil. O_2 concentrations were positively correlated with the earthworm biomass ($r = 0.69$, $p < 0.01$) and air-filled porosity ($r = 0.85$, $p < 0.01$), whereas CO_2 concentrations were negatively correlated with these parameters ($r_{earthworm} = -0.72$, $p < 0.01$, $r_{porosity} = -0.87$, $p < 0.01$).

The N_2O concentration was negatively correlated with air-filled porosity ($r = -0.61$, $p = 0.06$) and O_2 concentration (Table 1), and positively correlated with CO_2 concentration

(Table 1). The correlation with O_2 and CO_2 concentrations explained about 30% of the total variation in N_2O concentrations.

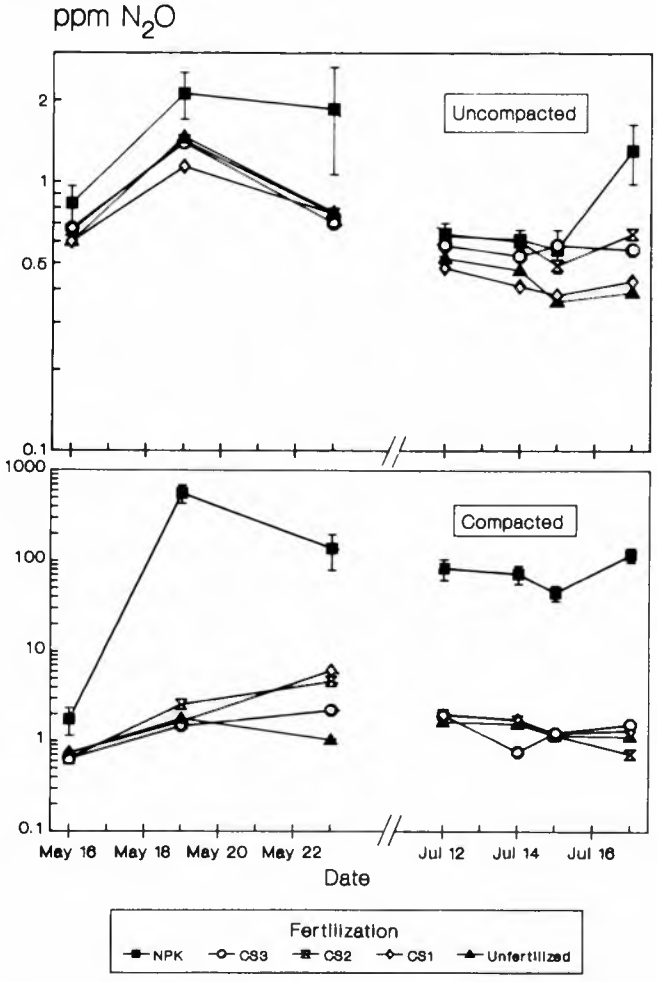

Fig. 2. N_2O concentrations in soil air (7-12 cm depth) with different soil compaction, cattle slurry (CS) and NPK fertilization (n = 4). The variability is indicated by standard error vertical bars on the NPK curve. The abbrevations are explained in material and methods.

Table 1. Correlation between N_2O concentrations in soil atmosphere (7-12 cm deep) and that of CO_2 and O_2 (Pearson correlation coefficients)[a] for the different treatments

	Uncompacted		Compacted	
Fertilization[b]	CO_2	O_2	CO_2	O_2
NPK, 126+94 kg N ha^{-1}	0.48*	-0.67***	0.50**	-0.60***
CS3, 117+72 kg N ha^{-1}	0.69***	-0.64***	0.66***	-0.75***
CS2, 78+53 kg N ha^{-1}	0.59***	-0.17ns	0.81***	-0.85***
CS1, 43+41 kg N ha^{-1}	0.71***	-0.50**	0.67**	-0.57**
Unfertilized	0.65***	-0.27ns	0.56**	-0.43*

[a] The correlation coefficients are based on 25-28 observations.
[b] NPK= NPK-fertilizer, CS = cattle slurry
***significant at 0.1% level, **significant at 1% level, *significant at 5% level, nsnon-significant.

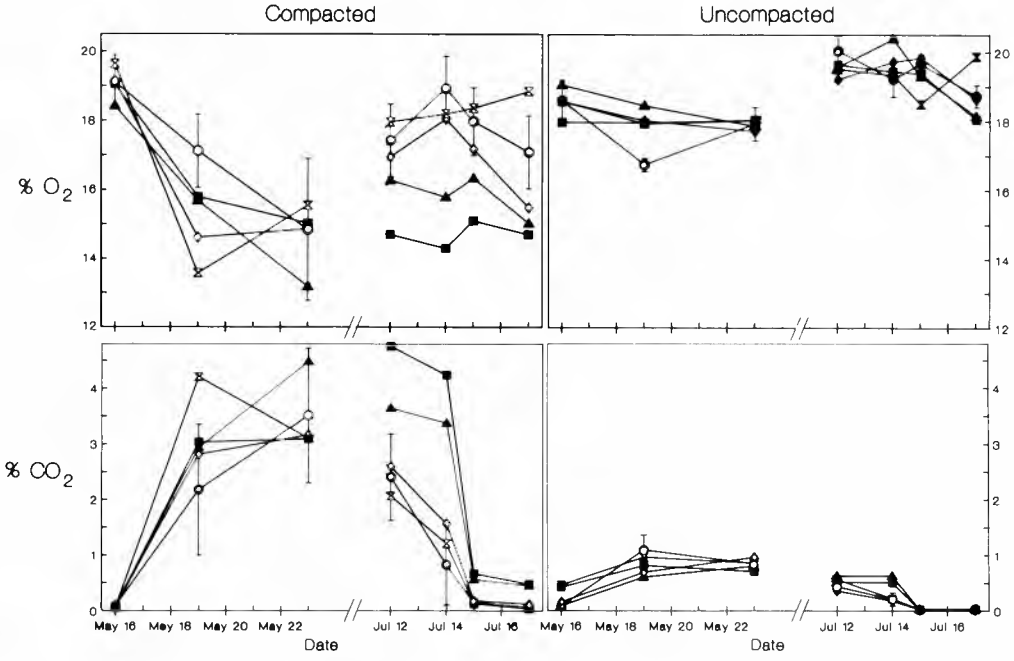

Fig. 3. CO_2 and O_2 concentrations in soil air (7-12 cm depth) with different soil compaction, cattle slurry (CS) and NPK fertilization (n = 4). The standard error is shown for the CS3 treatment (vertical bars). Symbols are the same as in Fig. 2

Mineral-N content in the soil was influenced both by the fertilization treatment and the soil compaction (Fig. 4). The N_2O concentrations in NPK-fertilized soil indicated that soil compaction induced large nitrogen losses through denitrification soon after fertilization. On this background, the NO_3 concentrations in these treatments early in the growth season were of particular interest as a tentative indicator of the denitrification losses. The measured NO_3 concentrations two weeks after fertilization were 3.57 μg NO_3-N g^{-1} soil dry matter in uncompacted NPK-fertilized plots and only 1.16 μg NO_3-N g^{-1} in the compacted ones. This difference (5 g NO_3-N m^{-2}) was taken as a tentative minimum estimate of nitrogen loss through denitrification induced by compaction, as discussed later.

In cattle slurry and unfertilized treatments NO_3 concentrations were low and there was no consistent effect of soil compaction. The amount of NO_3-N in later samples (23 June and 22 August) was low, and there were only minor differences between treatments.

N_2O concentrations on 19 May were positively correlated with NO_3 content measured one day later (20 May, $r = 0.61$, $p = 0.06$) in compacted treatments. In uncompacted treatments there was no statistically significant correlation between the same parameters.

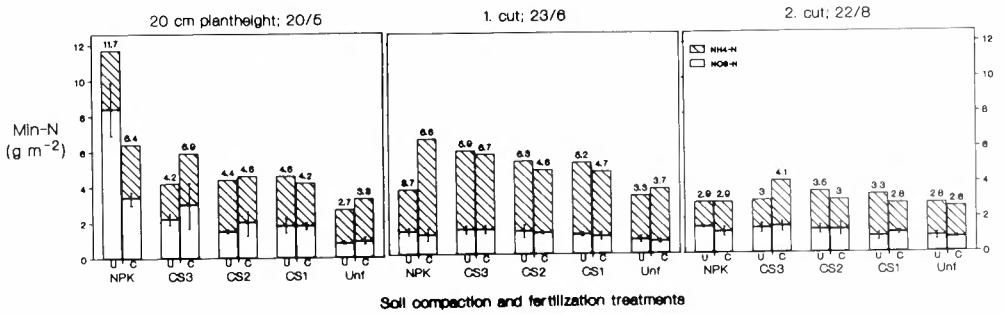

Fig. 4. Mineral nitrogen (g N m^{-2}) in soil (0-20 cm depth) with different soil compaction, cattle slurry (CS) and NPK fertilization (n = 2). The standard deviation is shown for the NO_3 concentration (vertical bars); U = uncompacted, C = compacted, other abbrevationes as in Fig. 2

DISCUSSION

The high N_2O concentrations in compacted NPK-fertilized, soil (Fig. 2) are consistent with the findings of Hansen *et al.* (1992) in a later growth season in the same field. During a period with heavy rain (June 1991), they found as much as 1900 ppm N_2O in soil air in a similar treatment. Preliminary investigations in the same field in 1988 (unpublished data, for three samplings taken 21, 34 and 48 days after fertilization) showed similar treatment effects, although the differences were less dramatic (3-4 times higher concentrations in the NPK plots than in the others). Maximum N_2O concentration in soil air reported in the present paper was 563 vpm N_2O (16 days after fertilization). Assuming a homogeneous concentration of N_2O throughout the 0-20 cm soil profile, 563 vpm is equivalent to about 580 g N_2O-N ha^{-1}. This is about 0.46% of fertilizer N added as NH_4NO_3 3 May (126 kg N ha^{-1}).

Lower N_2O concentrations in 1988 and 1989 compared with those in 1991 are probably a result of less precipitation, but losses during the storage must be considered. Klemedtsson (pers. com.) found venoject tubes to be suitable for N_2O storage. The highest CO_2 concentrations we found in 1989 were 25-100% of the maximum CO_2 concentrations found after very heavy rain in the same treatments in 1991 (Hansen *et al.* 1992). This indicates that the one year of frozen storage did not introduce gross errors in the CO_2 data. The N_2O data were lower, but within the same size. They had the same pattern as in 1991.

Textural change between the top layer and the gravelly subsoil may preclude efficient draining of the topsoil in this fluvial deposit (Hansen *et al.* 1992). Because of the high water content in the soil and the weak structure in this sandy loam, the soil structure is easily deteriorated by soil compaction. This will retard the gas diffusion through the soil layer. The retarded diffusion is clearly demonstrated by the large differences in O_2 and CO_2 concentrations in compacted versus uncompacted soil. Hence, the possibilities for anaerobic spots in soil will increase as a result of soil compaction. The frequency of anaerobic or microaerobic sites in the soil has clearly stimulated N_2O production. This is most clearly

seen in NPK-fertilized plots (Fig. 2). The high peak in N_2O concentration in soil air found in this treatment is probably caused by denitrification. N_2O represents only one fraction of the gaseous products of denitrification. In wet soils N_2 probably represents a larger fraction (Davidson 1991). As mentioned earlier, the soil compaction resulted in a substantial reduction in the NO_3 concentration in NPK-fertilized soil measured two weeks after fertilization (Fig. 4). This reduction is hardly caused by differences in the leaching of NO_3-N. Neither are the differences attributable to variations in plant uptake; the plant uptake of soil-N was reduced rather than stimulated by the soil compaction (Hansen 1992b). Hence, the difference of 5 g NO_3-N m^{-2} between compacted and uncompacted soil probably represents a minimum estimate of denitrification N-loss induced by soil compaction. Interestingly, the N-balance estimates (Hansen 1992b) indicate losses of the same magnitudes. The nitrogen input through fertilization, fixation and atmospheric deposition, exceeds output (cropped N) with 4 g m^{-2} in compacted but not in uncompacted soil. The difference might be ascribed to compaction stimulated denitrification. There is good agreement between these estimates (4 versus 5 g m^{-2}) considering the gross assumption made.

The 5 g m^{-2} represents 40% of the fertilizer nitrogen added 3 May. This is a very high loss compared to other findings (Eichner 1990, Rheinbaben 1990). A maximum nitrogen loss of 10% caused by denitrification is often observed, but larger losses are also reported by Rheinbaben (1990). He refers to denitrification estimates by the ^{15}N balance and acetylene inhibition methods ranging from 0 to 77% of total fertilized N. Rheinbaben expects larger denitrification losses after addition of organic carbon. He assumes carbon and oxygen to be the main regulating factors of denitrification. This does not seem to be the fact in our case, where we got a rapid effect on N_2O concentration after addition of NH_4NO_3 but not after addition of cattle slurry. This supports the findings of Klemedtsson *et al.* (1991) who found that NO_3 proved to be a useful variable in predicting denitrification rates in the grass ley. Our findings could be due to ample amounts of available carbon after the tractor's damaging effects on the ley plants (Zhezmer *et al.* 1990). Later soil compaction treatments in the same field experiment (1991) were carried out on bare soil before sowing, and this resulted in very similar pulses of N_2O in NPK-fertilized soil (Hansen *et al.* 1992). Thus, the available carbon in this soil seems in both cases to be sufficient to bring about large denitrification losses when the soil is compacted. A dual role of plant roots might be hypothesized: On one hand they serve as a carbon-source for microorganisms when they are destroyed by compaction, on the other hand the dense network of plant roots may make the soil more resistant to soil compaction.

When cattle slurry is added, denitrification has to be fed by NO_3 produced by nitrifying bacteria. This process will take some time, and will not give such large NO_3 concentrations in soil as those obtained by fertilization with NH_4NO_3 . In compacted soil fertilized with cattle slurry, the denitrification rate is therefore likely to increase throughout the growth period. This is indicated by the increase in N_2O emissions towards the end of the examination period in May in these treatments (Fig. 2). In uncompacted soil, the risk of denitrification is much lower due to better aeration and more efficient NO_3 uptake by the plants.

The larger denitrification as a result of soil compaction in NPK-fertilized soil than in cattle slurry fertilized soil was not reflected in the yields. In cattle slurry fertilized plots, the yields were on average reduced from 10.1 t dry matter ha^{-1} to 7.3 t ha^{-1} after soil

compaction, whereas in NPK-fertilized plots the yields were reduced from 12 t dry matter ha^{-1} to 10.3 t ha^{-1} (Hansen 1992a). This reflects other negative effects of soil compaction (Hansen 1992a,b).

In conclusion, NH_4NO_3 fertilization after tractor traffic in rainy conditions is likely to result in large nitrogen losses through denitrification. Tractor traffic is likely to stimulate denitrification because of restricted aeration, and NH_4NO_3 fertilization because of the addition of easily available NO_3-N.

ACKNOWLEDGEMENTS

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Ammonium and nitrate as sources of nitrogen for vegetable transplants

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Cabbage and tomatoes were raised in plastic pots with peat moss as the growing medium. Leaching was avoided by placing the pots inside plastic bags. The peat was limed and supplied with P, K, Mg and micronutrients. Nitrogen was added at three levels, using different commercial nitrogen fertilizers alone or in mixture. At the high level of N the best growth was obtained with ammonium nitrate or a combination of ammonium nitrate and potassium or calcium nitrate. At the low level of N, calcium nitrate or potassium nitrate gave results equal to those achieved with ammonium nitrate. An adverse effect of nitrate alone that occurred at the high N level, may have been due to salt stress. Ammonium N alone, and especially at the high level, damaged the plants.

Key words: Ammonium toxicity, nitrogen fertilizer, peat moss, vegetable transplants

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In Norway vegetable transplants are mostly raised in peat moss. The peat is normally supplemented with a complete fertilizer containing nitrogen as ammonium nitrate. This investigation was undertaken to find whether other sources of nitrogen might be preferable.
The experiments were supported by the Norsk Hydro A/S.

MATERIALS AND METHODS

The peat moss in the experiments was supplemented with: 6 g dolomitic limestone (to pH about 5.8), 1 g superphosphate (9 % P) and 200 mg Fritted Trace Elements No 36 per litre. Different nitrogen fertilizers were mixed into the peat at three levels:

N1 = 100-110 mg N per litre
N2 = 200-220 mg N per litre
N4 = 400-440 mg N per litre.

The nitrogen was added as potassium nitrate (KN), "Kalksalpeter" (calcium nitrate) from Norsk Hydro (= KS, 13.5 % NO_3-N, 1% NH_4-N), "Kalkammonsalpeter" (ammonium nitrate) from Norsk Hydro (= KAS, 27.6% N), ammonium sulphate (AS), or a mixture of two of the nitrogen sources (Table 1).
Potassium was added at the rate of 300 mg K/l as potassium nitrate (in A1 and A2) or as

potassium sulphate.

The experiments had 18 different treatments, as indicated in Table 1.

Table 1. Nitrogen fertilizer per litre peat moss

	N1	N2	N4
A1	0.8 g KN	0.8 g KN 0.7 g KS	0.8 g KN 2.1 g KS
A2	-	0.8 g KN 0.4 g KAS	0.8 g KN 1.2 g KAS
A3	0.7 g KS	1.4 g KS	2.8 g KS
A4	0.4 g KAS	0.8 g KAS	1.2 g KAS
A5	0.5 g AS	1.0 g AS	2.0 g AS
A6	-	1.05 g KS 0.25 g AS	1.75 g KS 0.75 g AS
A7	-	0.7 g KS 0.4 g KAS	0.7 g KS 1.2 g KAS

The experiments were carried out with tomato 'Virosa' and cabbage 'Lennox' in a greenhouse. The tomatoes were placed in a warmer compartment than that for the cabbage. The plants were seeded in plastic pots, 12 cm in diameter, with about one litre of loose peat added and compressed to 800-850 ml in the pots.

Two to three seeds were planted in each pot. A short time after emergence all seedlings except one were removed from each pot. Each plot had three pots, and there were three replicates in the experiments. The raising period lasted for 4-5 weeks.

Every pot was placed in a transparent plastic bag to avoid leaching of nutrients.

Three experiments with identical treatments were carried out. In Experiment 1, conducted in June-July 1991, the temperature was for most of the time higher than optimum for growth, especially for cabbage. On some days the growth was restricted by water stress. In Experiment 2, conducted in September-October 1991, the temperature in the greenhouse was optimal for raising transplants: 15-20°C for cabbage and 20-24°C for tomatoes. In Experiment 3, conducted in February-March 1992, the natural light was supplemented with light from high-pressure sodium lamps.

RESULTS AND DISCUSSION

In Experiment 1 the germination of cabbage was delayed at all N4 treatments, but most marked at A5N4. This effect might have been due to the high temperature and was less pronounced in the other experiments.

In all experiments, the plants grew faster with N1 treatments than with N2 and N4 at the beginning of the experimental period. At the end of the experiments, the plants were higher in fresh and dry weight with the N2 than with the N1 treatments (Table 2). This shift in response indicates that nitrogen deficiency gradually delayed growth with the N1 treatments. In Experiment 1 the best final result of cabbage was obtained with N4 because the plants were harvested later than those in Experiments 2 and 3.

At a high level of nitrogen, nitrate fertilizers delayed growth more than ammonium nitrate. This effect was most likely due to a higher level of soluble salts in the plots where all the nitrogen was supplied by nitrate as compared with ammonium nitrate.

On average, the best growth was obtained with A4 (ammonium nitrate), A2 (potassium nitrate + ammonium nitrate), A6 (calcium nitrate + ammonium sulphate) and A7 (calcium nitrate + ammonium nitrate).

Table 2. Results at the end of the three experiments: height of tomato plants in millimetres and fresh weight of tomato and cabbage plants in grams

Treatments	Tomatoes (mm)			Tomatoes (g)			Cabbage (g)		
	Experiment			Experiment			Experiment		
	1	2	3	1	2	3	1	2	3
A1N1	312	223	115	26.2	11.4	19.4	33.1	13.0	6.9
A1N2	353	223	102	38.2	12.1	18.4	54.7	18.6	7.8
A1N4	303	168	88	33.5	6.8	13.2	66.1	14.6	4.6
A2N2	361	235	103	39.2	13.0	20.0	58.0	19.7	7.1
A2N4	349	226	95	36.8	9.5	16.4	74.2	22.0	8.1
A3N1	294	235	104	24.4	13.5	18.9	31.9	13.3	6.7
A3N2	319	223	99	38.6	11.2	18.9	50.3	16.7	6.3
A3N4	317	201	81	32.2	8.3	11.4	73.0	15.5	7.4
A4N1	308	256	107	27.2	15.5	19.2	33.8	13.8	6.7
A4N2	323	232	105	37.1	12.8	20.4	61.1	22.9	7.1
A4N4	331	214	95	40.9	8.4	16.3	83.2	22.6	7.3
A5N1	281	250	96	23.3	13.7	15.8	30.1	13.2	3.6
A5N2	243	204	74	22.7	9.0	10.6	31.3	15.3	2.6
A5N4	163	161	62	10.6	7.1	5.9	14.0	10.7	1.7
A6N2	331	233	99	41.8	12.5	19.0	52.1	20.6	7.7
A6N4	318	180	92	39.4	7.2	15.7	75.1	19.6	8.3
A7N2	346	233	105	42.4	13.2	21.2	59.8	22.2	8.4
A7N4	296	203	99	35.5	8.7	17.6	78.0	19.4	6.5

When all the nitrogen was given as ammonium (A5), the growth was delayed. There may be three different reasons for the delay in growth brought about by a high level of ammonium N: low pH, mineral imbalance or ammonium or ammonia toxicity.

When all the nitrogen is supplied as ammonium, the pH in the medium or in the rhizosphere may drop below 5.0 and create problems associated with acid soils (manganese toxicity, and so on).

With a high supply of ammonium, the uptake of K, Ca and Mg will be reduced, which can result in mineral imbalance in the plants (Barker et al. 1988).

Barker et al. (1988) found that when all the nitrogen given to radish in sand culture was supplied as ammonium, the pH in the medium was 3.5. Stunting, yellow-green chlorosis and necrosis, assumed to be caused by ammonium toxicity, masked the symptoms of Ca deficiency. As the Ca concentration in the solution increased, this injury increased. The authors considered the damage to be caused by ammonium toxicity.

Ikeda & Osawa (1981) claimed that the preference in absorption between ammonium and nitrate was different among vegetables. In a solution culture they found that cabbage and tomatoes absorbed NO_3-N dominantly at pH 5.0, but absorbed both forms almost equally well at pH 7.0. When plants were cultured with ammonium N, a relatively close relationship was observed between growth response and leaf-NH_4^+ concentration. The concentration of NH_4^+ in leaves was high compared to the concentration of amid-N.

The results obtained by Ikeda & Osawa (1981) indicate that the adverse effects of ammonium sulphate at the N4 level in the present experiments could be due to toxic concentrations of ammonium in the plants. On the other hand, Marschner (1990) summarized that the toxicity of a high level of ammonium often results from ammonia (NH_3), which affects plant growth and metabolism at low concentration levels at which NH_4^+ is not harmful. But the NH_3 concentration in the soil solution is very dependent on the level of the pH. The pH in the peat in the present investigation was about 5.6, so it is not likely that ammonia toxicity could be responsible for the reduced growth.

In general, it has been found that plants may accept NH_4^+ as the only nitrogen source at low temperatures, while NH_4^+ can be harmful at high temperatures (Mengel & Kirkby 1987). Accordingly, the specific adverse effect of ammonium sulphate in Experiment 1 may be related to the high temperature in that experiment.

In the present experiments leaching of nutrients was avoided. In commercial production some leaching of nitrates will always take place when plants are raised in peat moss. Ammonium is partly absorbed to the surface of the organic colloids and is less prone to leaching. Accordingly, if nutrients are not continuously added with irrigation water, ammonium nitrate may give better results than calcium nitrate or potassium nitrate as a nitrogen source. In addition there is clearly less risk of salt stress when ammonium nitrate is mixed into the peat moss compared with using the same level of N as nitrate.

A practical conclusion is that when dry fertilizer is mixed into peat moss used for raising plants, ammonium nitrate, or a complete fertilizer containing N as ammonium nitrate, should be preferred.

When the plants are irrigated with a nutrient solution during the raising period, the optimal ratio between ammonium and nitrate will most likely be different. The results from investigations in solution culture indicate that the solution should have only 10-20 % of the nitrogen as ammonium N.

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Entomopathogenic nematodes found in Norway

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Soil samples were taken in Southern Norway to investigate the occurrence of entomopathogenic nematodes in the families Steinernematidae and Heterorhabditidae. Nematodes in the genus *Steinernema* were recovered from 18 % of the samples collected using the *Galleria* baiting technique. The survey showed that *Steinernema* spp. are found naturally in Norwegian soils, and appear to be most common in Vestfold. Heterorhabditid nematodes were not isolated in this study.

Keywords: Entomopathogenic nematodes, Norway, Steinernematidae, survey.

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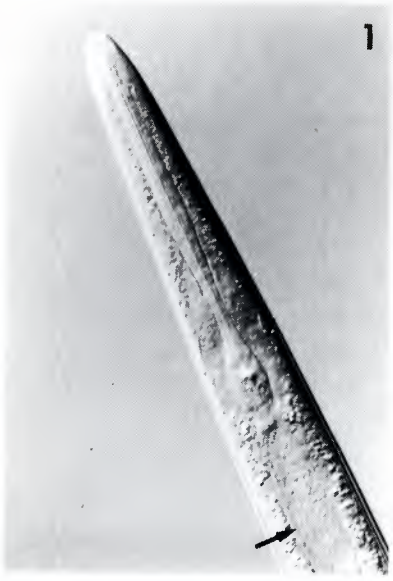
Entomopathogenic nematodes in the genus *Steinernema* and *Heterorhabditis* are parasites of a wide range of insect hosts, mainly soil-dwelling stages of the Lepidoptera, Coleoptera and Diptera. Their lifecycle includes a non-feeding third stage infective juvenile that carries symbiotic bacteria (*Xenorhabdus*) in its intestine. These infective stages are found naturally in the soil where they are attracted to suitable insect hosts; the nematodes enter through the natural openings of the host and once they have invaded the haemocoel through the mid-gut wall they release the bacteria. It is also known that the nematodes themselves release toxins to facilitate reproduction of the bacteria. Research is still required on this aspect however. The bacteria multiply rapidly causing septicaemia and death of the host after 24 to 48 hours, during which time the nematodes feed on the bacteria and reproduce within the cadaver. After the nutrients are depleted, infective stages are once again formed and leave the cadaver, to await a new host.

Several species of *Steinernema* and *Heterorhabditis* are currently being mass produced *in vitro* and sold as commercial biological control agents against certain insect pests.

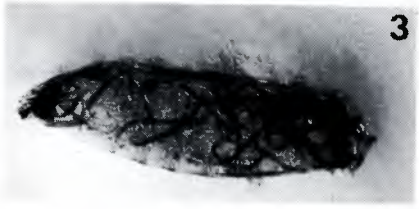
Isolation of these nematodes has been carried out successfully by using insect larvae, mainly *Galleria mellonella* (the greater wax moth), as bait to attract infectives from the soil. This method was developed by Bedding and Akhurst (1975), and surveys have shown that these nematodes are widely distributed in many parts of the world.

Steinernematid and Heterorhabditid nematodes have not been previously isolated in Norway, hence a survey was conducted to examine the occurrence in the southern part of the country, with the aim also of isolating nematodes for future use as biological control agents.

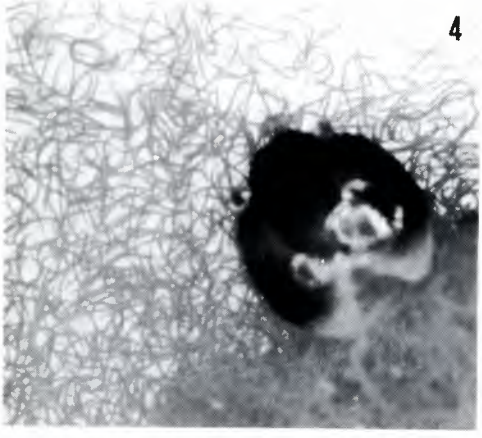


ANTERIOR END OF INFECTIVE THIRD STAGE JUVENILE.

2. INFECTION



REPRODUCTION WITHIN THE HOST.



INFECTIVE JUVENILES ARE FORMED AND LEAVE THE HOST CADAVER.

Fig. 1 General life-cycle of *Steinernema*

1. Non-feeding infective juveniles found naturally in soil, carrying the lethal symbiotic bacteria (arrow).
2. Under suitable environmental conditions, infectives enter a susceptible host through its natural body openings. Nematode releases nematode toxin(s) and bacteria, the host dies within 24 - 48 hours.
3. Reproduction of nematodes and bacteria occurs in the host, the former feeding on the latter. Adult nematodes increase greatly in size and one or two generations develop depending on available nutrients.
4. When nutrients in the cadaver are depleted (10-14 days at 24°C) infective juveniles are formed which then leave the host to search for or to await a new host.

MATERIALS AND METHODS

A total of 71 sites were sampled between May and October 1989. The three areas surveyed were Vestfold, Aust-Agder and Rogaland (Fig 2).

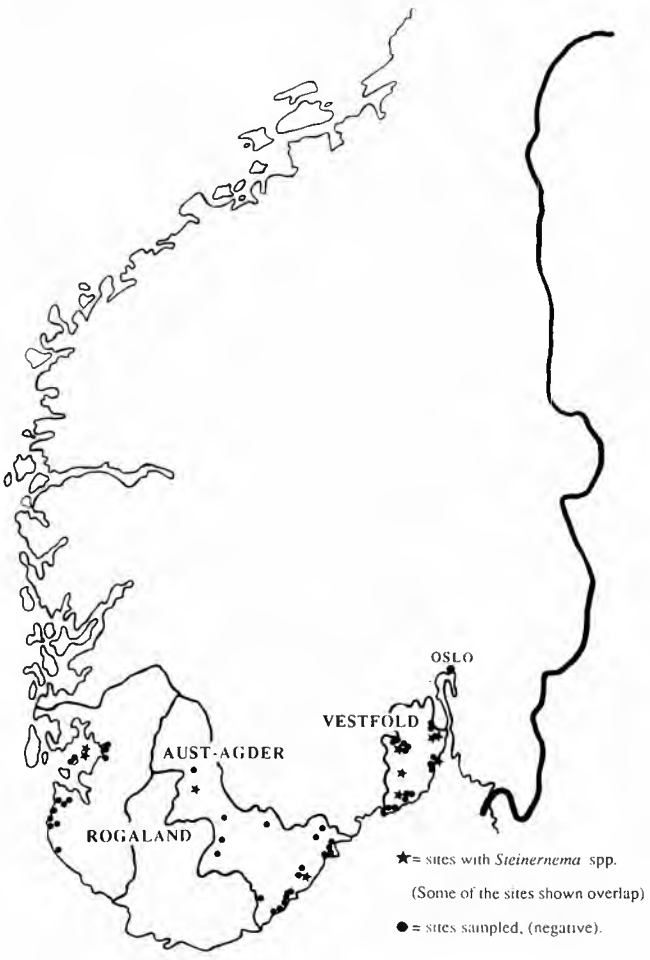


Fig. 2. Survey area showing soil sample sites

Soil samples were collected by growers or personally in the different areas. Information on soil type was provided by the growers and the vegetation was noted for each site. For each 100 m^2 site 10 samples were taken with a garden spade or soil corer down to a depth of about 15 cm and placed separately into 500ml plastic containers. A total of 710 samples were thus collected and placed in a cool dark place before analysis.

Prior to analysis samples were kept at room temperature for one week before two late instar larvae of *Galleria mellonella* were placed into each sample container as bait. After about one week each bait was examined, and any dead larvae were washed and dissected

to observe for possible nematode infection. Those *Galleria* that were infected were either left so that infective stages could develop for further culturing, or adult nematodes were placed on artificial medium (Dutky *et al* 1964), and infectives were collected and used to establish *in vivo* laboratory cultures. The baiting procedure was repeated once, to investigate whether there was an increase in infected *Galleria* larvae.

Morphological measurements were taken of infective stages and adult males for identification (Poinar 1986).

RESULTS

The overall occurrence of entomopathogenic nematodes in the survey was 13 positive sites out of 71 sampled (Table 1). All the nematodes isolated were in the genus *Steinernema*, and those identified to species were *S. feltiae* (= *bibionis*), *S. affinis* or *S. intermedium*. The species found in the different areas are presented in Table 2.

Table 1. Occurrence of *Steinernema* in the survey

Area sampled	Number of sites sampled	Number of sites positive (%)
Vestfold	28	9 (32)
Aust-Agder	25	2 (8)
Rogaland	18	2 (11)
Total Area	71	13 (18.3)

In a number of cases dead *Galleria* bait contained only a few adult nematodes that were often dead, but could nevertheless be identified to genus; however, these were not successfully cultured, hence not all the nematodes isolated from the various sites could be examined further. When nematodes were found they were usually the sole parasites apart from saprophytic nematodes and bacteria where the bait had deteriorated too quickly. In addition to nematodes, much of the *Galleria* bait was frequently found infected with entomopathogenic fungi, but this was not quantified, although some were identified as *Metarhizium anisopliae*, *Paecilomyces lilocinus* and *Aspergillus flavus*; the two former fungi are being kept in culture at Rothamsted Experimental Station.

The samples were baited twice, but showed little increase in the number of isolates found after this second baiting. For example, in the Vestfold area the first baiting gave an incidence of 25%, and after the second baiting an increase of 7%, giving a total incidence of 32%. The 10 samples taken from each site often contained more than one positive sample, and after a second baiting additional isolates were quite frequently found from the same site. This was true for Vestfold, whereas in Aust-Agder or Rogaland only one positive sample per site was observed, and a second baiting did not reveal any new positive sites. Most *Steinernema* isolates from Vestfold and one isolate from Aust-Agder have been successfully cultured on *Galleria* in the laboratory.

Table 2. *Steinernema* species found in the survey

AREA	LOCALITY (Grower)	POSITIVE SITES WITH *Steinernema* spp. (number of isolates found per site[1])		
		S. feltiae	*S. affinis and S. intermedium*	*Steinernema*. spp[2]
VESTFOLD	**Steinsholt** (T. Steinsholt)	1 (1)		1 (3)
	Åsgårdstrand (T. Stenersen)			2 (4)
	Tjøme (S. Fjellberg, Kjære)	1 (1)	1 (6)	
	Sem (O.Haugar,Broen)	1 (2)		
	Kvelde (N.Ø. Rimstad)	1 (3)		
	Larvik (Bokeskogen)			1 (1)
AUST-AGDER	**Kongshamn** (P. Holt, Alve)		1 (1)	
	Rysstad (Brokke)			1 (1)
ROGALAND	**Byre** (O. Byre)			1 (1)
	Skartveit (O. Halsne)			1 (1)

1) Within each site often more than one of the 10 samples contained *Steinernema* spp., these isolates are cultured seperately, even though they appear to be of the same species

2) These could only be identified to genus, since they represent *Galleria* bait that contained only a few adult nematodes that were sometimes dead

The distribution of soil types in the samples for the survey areas is shown in Table 3, and it can be seen that most soils sampled in Vestfold were sandy, followed by humic soils as well as some clay and moraine soils. In Aust-Agder and Rogaland most soils were loamy sands, and these two areas had more clay and less humic soils than samples from Vestfold. The frequency of *Steinernema* found in relation to soil type is shown in Table 4. Most nematodes were found in sandy or humic soils compared to loamy sands or clay and moraine soils. With regard to vegetation all areas represented similar biotypes i.e. mainly vegetable fields and orchards, woodland, pasture or meadows and grass or cereal fields, (Table 5).

As mentioned previously, most samples were taken between May and October. In Vestfold 7 sites were sampled in June of which 2 sites were positive (28.6%), the remaining 14 were taken in late October of which 7 were positive (33.3%). In Aust-Agder of the 9 sites sampled in May none were positive, of the 4 sampled in August one site was

positive (25%) and the 10 sites sampled in October also gave one positive site (10%). Finally, in Rogaland all sites were sampled in September of which 2 were positive (11%).

Table 3. Distribution of soil types in the three areas surveyed

Soil types	Distribution of soil types in survey areas (as % of number of sites for each area)		
	VESTFOLD	AUST-AGDER	ROGALAND
Sandy	53.6	8.0	-
Loamy sand	-	68.0	50.0
Clay	14.3	-	5.5
Clay loam	-	24.0	16.6
Humic	18.0	-	11.1
Morainic	7.1	-	11.1
Peat	-	-	5.5
Unknown	7.1	-	-

Table 4. Frequency of *Steinernema* according to soil type

Soil type	Number of sites with *Steinernema* (% of total)	
Sandy	4	(30.7)
Humic	4	(30.7)
Loamy sand	2	(15.4)
Clay loam	1	(7.7)
Clay	1	(7.7)
Moraine	1	(7.7)

Table 5. Types of habitats sampled

Types of habitats	VESTFOLD	AUST-AGDER (% of total number of samples)	ROGALAND
Vegetable crops	14 (50)***	15 (60)*	8 (44.5)*
Fruit (orchard/strawberries)	5 (18)*	7 (28)*	4 (22)*
Cereal (wheat/silage)	5 (18)**	ns	2 (11)
Pasture (incl. meadow)	2 (7)*	2 (8)	4 (22)
Forest	2 (7)**	1 (4) (Conifer nursery)	ns

*) sites with *Steinernema*
ns) habitat not sampled

Table 7 shows that average temperatures were close to normal in most cases for all the areas sampled. In all areas the rainfall differed somewhat from normal for that time of year. In Vestfold the rainfall in June was generally 59% of the norm, but in one area it was 85% close to normal. In October, however the rainfall was closer to normal around (70-86% near normal). For Aust-Agder rainfall was low, particularly in May when most places recorded only about 27% of the normal, whereas in August the rainfall was close to normal. In Rogaland the rainfall was below average, generally half of the norm, except near the coast, where rainfall was 80% close to normal.

Table 6. Soil types in the three areas as a whole from a soil map of Norway

AREA	PZ	BE	LT	SW	V/G	R
			% of soil types for area			
Vestfold	10-20	30.50	10-20	5-10	<5	-
Aust-Agder	30-50	10-20	20-30	5-10	<5	5-10
Rogaland	30-50	10-20	20-30	10-20	-	5-10

Abbreviations: PZ: Podzols, BE: Brown earths, LT: Lithosols, SW: Swamp soils, V/G: Vertic and gleyic cambisols, R: Ranker-like soils
After: Låg (1983) and Låg (1976)

Table 7. Temperatures and rainfall measured in the survey area

	VESTFOLD		AUST-AGDER		ROGALAND
	June	October	May	August	September
Average	15.5	7.6	11.1	14.3	11.1
Temperatures (°C)	(14.3)[1]	(6.6)	(9.7)	(16.1)	(11.2)
(nearest sites to	15.8	8.2	10.4	15	12.3
sampling for air	(14.8)	(7.5)	(9.7)	(16.1)	(12.5)
temperatures)					
			9.9	13.6	12.2
			(9.6)	(14.7)	(12.3)
Average rainfall	33	80	18	78	95
(mm)	(57)	(93)	(65)	(148)	(237)
(nearest sites to	44	84	13	59	95
sampling for rainfall	(74)	(119)	(48)	(100)	(145)
measurements)					
	61	88	19	132	98
	(72)	(105)	(75)	(155)	(122)
			19	105	
			(47)	(115)	

[1]) Figures in parentheses show normal temperatures and rainfall for that month

DISCUSSION

Steinernematid nematodes were most frequently found in Vestfold compared to Aust-Agder and Rogaland, (Table l). The apparent higher occurrence in Vestfold may be related to the lighter sandy soils that predominate this area, although in Aust-Agder and Rogaland a fair proportion of the soil was loamy sand. A soil map of Norway (Table 6) also indicates differences in soil types for the three areas as a whole. Most notably, Vestfold has a larger proportion suitable for cultivation (brown earths) compared to Aust-Agder and Rogaland, which have higher proportions of podzols and lithosols. However, the presence of suitable hosts and other unknown ecological or environmental factors could account for the differences in the incidence found. Such variations in prevalence among the different areas sampled have also been observed in surveys conducted in Great Britain and the Republic of Ireland (Hominick & Briscoe 1990, Griffin *et al* 1991), where some areas gave an incidence well below or above the overall average. Reasons for these differences were attributed to an association between recovery of nematodes and soil type, where light porous soils appeared to be the most favourable, and, interestingly, calcareous soils (Hominick & Briscoe 1990). In this study most Steinernematids were found in either sand or humus soil types, (Table 3), whereas a few were found in loamy sand, moraine and even clay. The results indicate, as reported by Hominick & Briscoe (1990) and Griffin *et al* (1991), that there seems to be an association between soil type and frequency of Steinernematid recovery.

In Vestfold *Steinernema* was recovered from all the habitats represented, and, in relation to the number of sites represented, forests sites contained *Steinernema* most frequently. If selective sampling had been conducted, more isolates might have been found. For the other two areas, *Steinernema* was isolated from vegetable field and fruit/orchard sites, but no nematodes were found in cereal or pasture sites. This is perhaps not surprising for Aust-Agder, where almost 90% of the samples came from vegetable and fruit sites. In Rogaland, habitat representation was a little more even and in this case the soil type may have been one of the limiting factors for *Steinernema* recovery. For both these areas forest sites were not sampled. *Steinernema* may be more frequently found in habitats where insect hosts are likely to be present; habitats such as forests and orchards or strawberry plants (which were well represented) would be favourable in this respect. *Steinernema* was more common in Vestfold, perhaps because of the habitats represented coupled with favourable soil type and near normal rainfall at the time of the survey. In conclusion, there appears to be no clear association between habitat and nematode recovery, although it is felt from the results that orchards/fruit fields and forest sites may be the most favourable for *Steinernema* recovery.

According to several surveys of Steinernematids and Heterorhabditids, there is some evidence of seasonal occurrence. The seasonality appears to follow a similar pattern where a decline in prevalence occurs in spring/early summer followed by a progressive increase towards late summer/autumn (Mracek 1980, Blackshaw 1988, Hominick & Briscoe 1990, Griffin *et al* 1991). In the present survey, samples were taken between May and October at different times for the three areas. If the two sampling periods in Vestfold are compared, there appears to be little difference in prevalence between early summer and late autumn; however in Aust-Agder samples taken in spring were negative compared with those taken in autumn.

Steinernema was the only genus found in the survey, which is perhaps not surprising since results from surveys in northern Europe also show that this genus is commonly found, particularly *S. feltiae*, *S. intermedium* and *S. affinis*. The *Heterorhabditis* species seems to be less frequently found and has not yet been reported in Scandinavia (present survey, Burman *et al* 1986, Husberg *et al* 1988), whereas it has been found in one site in Great Britain and another in the Republic of Ireland (Hominick & Briscoe 1990, Griffin *et al* 1991).

As one goes further south, to for example, a survey in Italy, 36% of the samples contained *Heterorhabditis* (Griffin *et al* 1991). This suggests that conditions in North Western Europe with its colder climate may limit the distribution of *Heterorhabditids*.

S. feltiae, which seems to be more prevalent in North Western Europe, may also tolerate the climatic conditions in this area better than, for example, *S. carpocapsae*, which appears to be less common. Although *S. carpocapsae* has been found only once in Great Britain (Georgis & Hague 1980), its known distribution is widespread, and it has been found in Europe, North America, South America, Australia and New Zealand. *S. feltiae*, however, has only been found in Europe, Australia and New Zealand, and it is thought that it may have been imported by man to the two latter areas, (Poinar 1990). In temperature studies comparing *Heterorhabditis* sp., *S. feltiae* and *S. carpocapsae*, the range at which these nematodes can develop within *Galleria* differs in that *S. feltiae* favours a range between 6 and 27°C, *S. carpocapsae* between 14 and 32°C and *Heterorhabditis* sp. between 16 and 27°C (Haukeland *et al* 1992). These results support the view that *S. feltiae* appears to tolerate lower temperatures and hence the reason for its general presence in North Western Europe. The apparent lower prevalence of *S. affinis* and *S. intermedium* compared to *S. feltiae* may be due to the latter species, higher capacity for survival, speculatively based, for example, on its ease of *in vivo* culture and survival of infective juveniles (personal observation), which was also noted by Griffin *et al* (1991).

The survey has shown that Steinernematids appear to be common in some parts of southern Norway; however, a limited number of samples were taken from a large area over a short period of time, thus giving only an idea of the distribution, and species or genera of nematodes present, at that particular time. Further repeated sampling at different times of the year, and more extensive sampling would increase the knowledge about their ecology and species or genus representation in the country.

The nematodes obtained from the survey are at present being used for research on biology and are currently being tested as biological control agents at the laboratory level. It is hoped that in the future these nematodes can be developed as successful biological control agents against susceptible pests in Norway.

SUMMARY

Entomopathogenic nematodes in the family Steinernematidae were found in southern Norway, most frequently in Vestfold. The nematodes were isolated from soil samples taken in Vestfold, Aust-Agder and Rogaland from May to October 1989. The most common species found were *Steinernema feltiae*, *S. affinis* and *S. intermedium*. Nematodes in the family Heterorhabditidae were not found in this survey. The frequent occurrence in

Vestfold may be attributed to favourable soil types in that region. The nematodes have potential for future use as biological control agents in the country, particularly *S. feltiae*.

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Breeding pears adapted to Scandinavian growing conditions: Screening of 46 advanced pear selections 1985-91

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In a screening test of 46 advanced pear selections from the Balsgård breeding programme the selections were compared to the standards 'Keiserinne' (syn. Epargne) and 'Moltke'. Some new selections were found to be outstanding in one or more characters. Characters assessed included yield, fruit size, fruit quality, shelf life, storage capacity, resistance to scab (*Venturia pirina*), bugs (*Miridae* and *Pentatomidae*) and fruit cracking, along with phenological data. The most promising candidates to become new cultivars were the selections BP 10273, BP 8565, BP 9357, BP 9627 and BP 10104.

Key words: Cultivars, pear, *Pyrus communis* L., selection

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New pear cultivars adapted to Scandinavian growing conditions have been rare in recent years, and only a few have been introduced to the Norwegian fruit industry in the present century. The most recent cultivars of importance are Herzogin Elsa and Précoce de Trévoux, both introduced in the late 1920s (Kvåle & Skard 1958). New cultivars have been tested, but none have shown any commercial value (e.g. Hjeltnes & Husabø 1989). Through a joint Swedish-Norwegian pear-breeding project carried out at The Swedish University of Agricultural Sciences, Balsgård, Department of Horticultural Plant Breeding, in cooperation with The Norwegian State Agricultural Research Stations, pear seedlings selected at Balsgård have been evaluated and reselected at Ullensvang Research Station at two locations, Njøs and Ullensvang, Western Norway (Nybom 1990).

MATERIAL AND METHODS

Scion wood of 46 pear selections was grafted or budded at Ullensvang Research Station, substation Njøs in the years 1983 to 1985, depending on the year of selection in the field at Balsgård. Two of the main cultivars in the Norwegian pear industry 'Keiserinne' and 'Moltke' were chosen as standards. By planting in May 1985 some selections were one-year-old maiden trees without feathers, while others were two-year-old trees with branches. The experiment had a randomized block design with one tree per plot spaced at 5 x 3 m. Three replicates were placed at Njøs and two at Ullensvang. Cultural practices were as standard in the district.

Field observations included date of first flower, harvest date, yield and number of fruits per tree. Fruits were evaluated after harvest according to damage caused by biotic or abiotic factors in the field, such as pear scab (*Venturia pirina*), bugs (*Miridae, Pentatomidae,* a.o.) and fruit cracking. This evaluation was made in 1990 and 1991 at Njøs and in 1990 at Ullensvang. At Njøs no fungicides or pesticides were applied during the last two years of evaluation. Fruits were stored at -0.5 to -1.0 °C and removed from the cold storage at different intervals. The fruits were ripened at 20°C for seven days and a sensorial test of fruit quality was carried out. After a ripening period of 14 days at 20°C a new test was carried out. All dates were recorded in Julian days.

The sensorial test was carried out by a panel of two persons at Njøs and two persons at Ullensvang. A 0-9 scale was applied, where 9 was excellent, 5 was acceptable and a score of 1 was regardes as very bad. The score zero was set if the fruits had completely deteriorated and could not be tasted.

In addition to the sensorial test, a record for "brown core" was taken at Njøs at each date of evaluation. Ten fruits were split longitudinally and evaluated on a scale of 0-9, where 0 indicated no sign of internal breakdown and 9 indicated complete deterioration. These records were noted on individual fruits, and the mean score of the selection on the specific day was kept for further analysis.

All statistical analyses were carried out by means of SAS (Statistical Analysis System).

The selections in the test are presented in Table 1.

RESULTS

The most important characters when selecting a new pear cultivar are phenological observations like flowering and harvest dates, yield capacity, fruit size, resistance to biotic and abiotic factors, shelf life and fruit quality.

Phenological observations

Harvest date

The date of harvest revealed that the earliest selections were harvested about the same time as 'Keiserinne', while the latest ones were harvested 20 days after 'Moltke'. Most selections were harvested in a four-week period prior to and after 'Moltke'.

Date of first flower

The date of first flower was found to be very early according to normal blossom time in Western Norway. This was due to a very early spring in two of the three years of observation. Both early and late flowering selections were identified, and the range was from 20 April to 12 May.

Table 1. Pear selections from Balsgård, tested at Ullensvang Research Station and substation Njos, 1985-91

BP 648	Colorée de Juillet	x Conference
BP 704	Belle lucrative	x Doyenne du Comice
BP 895	Herzogin Elsa	x Belle lucrative
BP 6517	Belle lucrative	x Doyenne du Comice
BP 6963	Conference	x Williams
BP 8039	Conference	x Clapp's Favorite, red
BP 8096	Herzogin Elsa	x Conference
BP 8405	Worden Seckel	x Fertility (2x)
BP 8565	Clapp's Favorite	x Conference
BP 8627	Clapp's Favorite	x Comtesse de Paris
BP 8640	Clapp's Favorite	x Comtesse de Paris
BP 8881	Conference	x Herzogin Elsa
BP 8892	Conference	x Madame Treyve
BP 9066	Marguerite Marillat	x Bonne Louise
BP 9071	Marguerite Marillat	x Belle lucrative
BP 9105	Colorée de Juillet	x Clapp's Favorite
BP 9171	Conference	x Williams
BP 9238	Worden Seckel	x Belle lucrative
BP 9292	Herzogin Elsa	x Packham's Triumph
BP 9328	Herzogin Elsa	x Flemish Beauty
BP 9357	Packham's Triumph	x Herzogin Elsa
BP 9431	Johantorp	x Carola
BP 9452	Doyenne de Juillet	x Clapp's Favorite
BP 9457	Colorée de Juillet	x Clapp's Favorite
BP 9478	Colorée de Juillet	x Clapp's Favorite, red
BP 9531	Dr. Jules Guyot	x Conference
BP 9540	Dr. Jules Guyot	x Conference
BP 9541	Dr. Jules Guyot	x Conference
BP 9627	Herzogin Elsa	x Packham's Triumph
BP 9667	Herzogin Elsa	x Beurré Superfin
BP 9676	Herzogin Elsa	x Le Lectier
BP 9803	Johantorp	x Carola
BP 9851	Colorée de Juillet	x Gränapäron
BP 9861	Beurré Giffard	x Helmershus
BP 10015	Clapp's Favorite	x Bonne Louise
BP 10022	Clapp's Favorite	x Bonne Louise
BP 10033	Clapp's Favorite	x Bonne Louise
BP 10036	Clapp's Favorite	x Bonne Louise
BP 10101	Herzogin Elsa	x Conference
BP 10104	Herzogin Elsa	x Conference
BP 10174	Herzogin Elsa	x Bonne Louise
BP 10201	Herzogin Elsa	x Bonne Louise
BP 10273	Conference	x Bonne Louise
BP 10529	Clapp's Favorite	x Soldat Laboureur
BP 10693	Clapp's Favorite	x Belle lucrative
BP 11014	Belle lucrative	x Carola
Moltke		
Keiserinne (syn. Epargne)		

Secondary flowering
The observations of secondary flowering at Njøs in 1989 revealed that most of the cultivars did not express this phenomenon in this specific year. However, the selection BP 9676 had an extremely high frequency of secondary flowers, and even the selections BP 6517, BP 8640, BP 9457 and BP 9531 had rather high numbers of secondary flowers.

Yield and fruit size

The yield capacity and fruit size were very different between the selections. It can be seen from Table 2 that the accumulated yield ranged from less than 0.1 to 42.0 kg per tree, and fruit size from 44 to 159 g on average. The most productive selections were BP 895, BP 10529, BP 8881, BP 9328, BP 8405, BP 10201 and BP 8627 in descending order, all exceeding 'Moltke'. However, only the first two selections had a significantly higher yield than 'Moltke'.

As far as fruit size is concerned, for the most productive selections only BP 10529 and BP 9328 had a fruit weight exceeding 100 g. This size should be reached provided the selections are outstanding in all other characters to become a promising selection for commercial production. Fruits of 'Keiserinne' averaged 74 g, and this small-fruited variety is kept because of its outstanding fruit quality and earliness. No other selection had a significantly higher fruit weight than 'Moltke'; however, BP 10273 had very large fruits at Njøs (183 g).

In the analysis of variance, location and replication were regarded as random effects, while selection was regarded as a fixed effect. Location x selection interaction was found to be significant both for yield and fruit size.

An analysis of variance for each individual selection indicated that the selections BP 9292, BP 10104, BP 9066, BP 9851, BP 9238, 'Keiserinne' and BP 10036 had significantly higher yields at Njøs, while the selections BP 6963 and BP 648 had significantly higher yields at Ullensvang. Fruit size was larger at Njøs for the selections BP 10529, BP 704, BP 10273, BP 9066, BP 9238, BP 9541, BP 8892, BP 6963, BP 9676 and BP 9540. As a mean for all selections, the accumulated yield per tree was larger at Njøs than at Ullensvang, i.e. 17.9 kg as compared with 15.9 kg. Fruit size was larger at Njøs, too, 111 g as compared with 96 g.

Resistance to biotic and abiotic factors

The results were variable because of differences between years and locations. Lack of yield in some cultivars in one year and/or location complicated the evaluation even further. Hence, no tabulation is made for resistance observations.

Resistance to bugs (Miridae, Pentatomidae a.o.)
No significant differences were found between selections in the incidence of this type of damage. The incidence was very low at Njøs; at Ullensvang, however, some damage was observed, but the data are too scarce to draw any conclusions.

Resistance to pear scab (Venturia pirina)
The incidence of pear scab (*Venturia pirina*) was much higher at Njøs than at Ullensvang mainly because no fungicide treatments were carried out at Njøs during the years of registration. At Ullensvang scab infection was noted for the selection BP 10201 exclusively, and this selection was highly susceptible at Njøs as well. Other selections that showed a high incidence of scab were BP 9627 and BP 10529. No other selection exceeded 10% damage by pear scab.

Table 2. Phenological observations, yield and fruit size in pear selections, in descending order of harvest date

Selection	Date of harvest	Date of 1st flower	Amount of secondary flowers	Accumulated yield per tree	Mean fruit size
BP 9851	240	123	1	21.5	44
Keiserinne	242	121	0	19.1	74
BP 648	243	128	0	16.8	98
BP 9452	246	131	1	9.8	100
BP 9238	250	123	0	20.9	115
BP 9457	250	126	2	8.7	128
BP 9328	252	125	0	34.1	126
BP 9478	252	122	1	4.9	130
BP 9861	253	129	1	12.5	108
BP 895	255	119	1	42.0	58
BP 9431	257	125	0	17.8	71
BP 10273	259	121	0	24.2	159
BP 9105	260	127	0	8.5	126
BP 9540	261	127	1	8.0	98
BP 9541	261	124	1	19.8	106
BP 8640	262	126	2	23.6	132
BP 8627	264	125	0	26.9	79
BP 8881	264	125	0	35.2	81
BP 9066	264	125	1	21.2	141
BP 9071	265	127	0	15.3	117
BP 10529	265	132	0	41.6	107
Moltke	266	125	1	26.6	149
BP 704	268	127	0	24.7	92
BP 8405	268	122	0	30.5	96
BP 10104	270	118	0	25.5	103
BP 10201	270	129	1	27.9	88
BP 10693	270	132	0	4.7	123
BP 11014	270	127	0	0.1	99
BP 10022	270	129	0	4.8	93
BP 8892	271	119	1	19.3	106
BP 9531	272	122	2	19.8	140
BP 10174	274	127	0	1.5	101
BP 9803	274	130	0	< 0.1	125
BP 6963	275	125	0	17.6	80
BP 9292	275	129	1	26.6	116
BP 10033	276	131	0	0.2	140
BP 9171	277	122	0	13.0	84
BP 9667	278	126	1	24.4	89
BP 6517	279	129	2	9.7	116
BP 10101	280	123	0	4.8	109
BP 8565	280	126	0	18.6	124
BP 9676	280	129	7	11.7	115
BP 8096	281	126	0	12.3	113
BP 10036	281	127	0	5.0	89
BP 8039	282	129	0	4.3	150
BP 9627	282	122	1	16.6	130
BP 10015	283	131	1	0.4	113
BP 9357	286	126	1	23.8	117
Mean				16.8	106

All dates are given in Julian days. Yield is the accumulated yield from 1987 to 1991 per tree.

Resistance to fruit cracking
The selections differed at the two locations in respect of fruit cracking. BP 10201 presented a high incidence of fruit cracking at Ullensvang, BP 9452 at Njøs while the opposite selection-location combinations were undamaged. Other selections with a high incidence of fruit cracking were BP 8039 and BP 9066 along with the cultivar 'Keiserinne'. Most other selections were resistant to this type of damage.

Fruit quality, shelf life and storage capacity

The present study was carried out on a wide range of genotypes and subsequent variability in yield. Hence the number of evaluations per genotype has been highly variable because of lack of fruits in some genotypes. Some selections have been harvested at non-optimal maturity, and the scores for shelf life and storage capacity are encumbered by some uncertainty.

Fruit quality
In Table 3 the scores for fruit quality for successive months are presented. In August very few notes were taken on fruit quality. No selection was as good as the standard 'Keiserinne'.
In September 'Keiserinne' had low scores. During the last week of September, when the evaluations were carried out, the optimum marketing period for this cultivar is passed. The best selections in September were BP 704, BP 895 and BP 10529. The general scores were rather low in this month, partly because of lack of sufficient ripening in some of the selections, partly because of few selections being evaluated.

In October the standard 'Moltke' gained high scores. Even higher scores were observed for the selections BP 10273, BP 10201, BP 9357, BP 8565 and BP 10104 in descending order. October was the month with the highest score for fruit quality during the pear season.

In November it was found that none of the selections averaged the score for acceptable quality over years, and none of the selections had a higher score than Moltke. The poor result for this month is mainly due to lack of sufficient data, but the strict selection connected to the pretreatment of the fruits is also a contributory factor. When making a comparison with the fruit quality data in December, the selections BP 8565, BP 9357, BP 10104 and BP 10273 had acceptable fruit quality, and it could be assumed that all these selections would most likely have acceptable fruit quality in November as well. In January good fruit quality was noted for the selection BP 9627 exclusively.

Shelf life and storage capacity
It can be seen from Table 4 that shelf life became shorter with increasing length of storage. At a defined period of ripening, both frequency and severity of "brown core" increases by prolonged storage. The early-maturing cultivar 'Keiserinne' had a low storage potential, and the comparable selection BP 648 had a slightly better shelf life. 'Moltke' performed well in September, but in succeeding months two weeks' ripening gave too high an incidence of "brown core" in this cultivar. Groups of selections with a better, equivalent or poorer shelf life than 'Moltke' were found in all months from October to January. The best selections in means over these months were BP 6963, BP 9627, BP 9676 and BP 10101. These selections were edible with respect to "brown core" even after 14 days of ripening in January.

Table 3. Mean scores for pear fruit quality in different months. Seven days' ripening at 20°C. Means of two or more assessments over years and location

Selection	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.
BP 648	4.2	2.5	3.9	.	.	.
BP 704	.	6.7	5.4	3.7	3.1	.
BP 895	.	5.7	4.5	3.9	3.1	.
BP 6517	.	.	.	4.2	4.0	0.0
BP 6963	.	.	2.2	2.8	3.1	4.7
BP 8039	.	.	5.0	.	.	.
BP 8096	.	.	5.3	4.7	4.4	3.2
BP 8405	.	4.5	4.5	4.3	3.4	.
BP 8565	.	.	6.0	4.2	6.3	.
BP 8627	.	3.5	3.9	3.8	4.4	.
BP 8640	.	3.7	4.8	4.8	2.8	.
BP 8881	.	4.0	3.9	4.5	4.3	.
BP 8892	.	3.0	4.9	4.0	4.8	4.2
BP 9066	.	2.2	3.2	2.8	2.7	.
BP 9071	.	.	2.9	3.0	1.0	.
BP 9105	.	2.2	2.9	0.0	.	.
BP 9171	.	.	3.9	3.9	3.1	.
BP 9238	.	.	3.7	3.2	3.2	.
BP 9292	.	.	5.0	4.0	3.7	.
BP 9328	.	4.0	5.0	3.5	4.3	.
BP 9357	.	.	6.0	4.0	5.7	.
BP 9431	.	.	3.8	4.3	3.6	.
BP 9452	.	.	3.5	.	2.8	.
BP 9457	.	.	3.1	2.2	1.0	.
BP 9478	.	.	5.0	.	0.0	.
BP 9531	.	4.2	3.5	3.2	2.4	0.8
BP 9540	.	.	3.6	2.7	3.0	.
BP 9541	.	4.5	2.9	1.3	0.0	.
BP 9627	.	.	3.7	4.4	4.4	6.0
BP 9667	.	.	4.9	3.6	4.3	0.0
BP 9676	.	.	4.5	4.5	4.0	4.7
BP 9803	.	.	.	.	.	.
BP 9851	3.7	4.2	3.0	.	1.5	.
BP 9861	.	4.7	3.0	1.5	2.0	.
BP 10015	.	.	.	.	.	.
BP 10022	.	.	.	1.8	.	0.0
BP 10033	.	.	.	.	.	.
BP 10036	.	.	.	4.5	2.8	.
BP 10101	.	.	.	3.7	3.7	.
BP 10104	.	4.7	5.7	2.7	5.3	1.5
BP 10174	.	.	.	.	.	.
BP 10201	.	4.0	6.0	3.5	3.8	.
BP 10273	.	3.7	6.7	4.2	6.2	.
BP 10529	.	5.5	4.3	4.5	4.5	1.5
BP 10693	.	.	5.0	4.5	3.3	.
BP 11014	.	.	.	.	.	.
Moltke	.	.	5.5	4.8	4.2	.
Keiserinne	7.0	4.3	3.7	.	0.0	.

Table 4. Shelflife in different months. Analyses carried out at 7 and 14 days' ripening at 20°C

	Aug.		Sept.		Oct.		Nov.		Dec.		Jan	
Selection	7	14	7	14	7	14	7	14	7	14	7	14
BP 648	+	+	+	-	+	-	.	.	.	.	.	.
BP 704	.	.	+	.	+	-	-	-	-	-	-	-
BP 895	.	.	+	-	+	-	+	-	-	-	.	.
BP 6517	.	.	.	.	.	.	+	+	+	-	-	-
BP 6963	.	.	.	.	+	+	+	+	+	+	+	+
BP 8039	.	.	.	.	+	.	.	.	.	.	.	.
BP 8096	.	.	.	.	.	.	+	-	+	-	-	-
BP 8405	.	.	+	+	+	+	+	-	-	-	-	-
BP 8565	.	.	.	.	+	+	+	-	+	-	+	-
BP 8627	.	.	+	+	+	+	+	-	+	-	.	.
BP 8640	.	.	+	+	+	-	+	-	+	-	-	-
BP 8881	.	.	+	+	+	+	+	-	+	-	-	-
BP 8892	.	.	+	+	+	+	+	-	+	-	-	-
BP 9066	.	.	+	-	+	-	+	-	-	-	.	.
BP 9071	.	.	.	.	-	-	-	-	-	-	-	-
BP 9105	.	.	+	.	-	-	-	-	.	.	.	.
BP 9171	.	.	.	.	+	+	+	-	+	-	-	-
BP 9238	.	.	.	.	+	-	+	-	-	-	.	.
BP 9292	.	.	.	.	+	+	+	-	-	-	-	-
BP 9328	.	.	+	.	+	-	+	-	-	-	.	.
BP 9357	.	.	.	.	+	+	+	+	+	-	-	.
BP 9431	.	.	.	.	+	+	+	+	+	-	.	.
BP 9452	.	.	.	+	-	-	-	-	-	-	.	.
BP 9457	.	.	+	.	-	-	-	-	-	-	.	.
BP 9478	.	.	.	.	-	-	-	-	-	-	.	.
BP 9531	.	.	+	+	+	+	-	-	+	-	-	-
BP 9540	.	.	+	.	+	-		-	-	.	.	.
BP 9541	.	.	+	-	-	-	-	-	-	-	.	.
BP 9627	.	.	.	.	+	+	+	+	+	+	+	+
BP 9667	.	.	.	.	+	+	+	-	+	-	-	-
BP 9676	.	.	.	.	+	+	+	+	+	+	+	+
BP 9803	.	.	.	.	.	.	.	.	.	.	.	.
BP 9851	+	+	+	-	+	-	-	-	-	-	.	.
BP 9861	.	.	-	-	-	-	-	-	-	-	.	.
BP 10015	.	.	.	.	.	.	.	.	.	.	.	.
BP 10022	.	.	-	-	-	-	-	-	-	-	.	-
BP 10033	.	.	.	.	.	.		.	.	.	.	.
BP 10036	.	.	.	.	.	.	-	-	-	-	-	-
BP 10101	.	.	.	.	.	.	+	+	+	+	+	+
BP 10104	.	.	+	+	+	.	+	-	+	-	-	-
BP 10174	.	.	.	.	.	.	+	.	.	.	.	.
BP 10201	.	.	+	+	+	-	-	-	-	-	-	-
BP 10273	.	.	+	+	+	-	+	-	-	-	.	.
BP 10529	.	.	+	+	+	-	+	-	+	-	-	-
BP 10693	.	.	.	.	+	-	+	-	+	-	-	-
BP 11014	.	.	.	.	.	.	.	.	.	.	.	.
Moltke	.	.	+	+	+	-	+	-	+	-	-	-
Keiserinne	+	.	+	-	-	-	-	-	-	-	.	.

"Brown core" is rated on a 0-9 scale, where 0 indicates no damage. Selection scores for "brown core" of < 3 are rated '+', scores of >=3 are rated '-', missing values are rated '.'

DISCUSSION

The present experiments have identified some promising new selections in pear, especially with regard to finding good autumn and winter cultivars. Even though the experiments were run for a short period of time, and many of the selections had too few fruits for a proper assessment of their value, the material is sufficient for drawing some conclusions. If the precocious yield capacity is not sufficient to provide an estimate for fruit quality, shelf life and resistance, the selection should not be considered as good enough for further trials. Acceptable precocity should be chosen by comparing with the bearing habit of the standards. Because of the lack of uniformity in the plant material, some of the selections in the experiments have been regarded as promising even though they have had too low yields.

It was found that new selections exceeded the standards for almost all recorded characters. When naming new cultivars however, one must try to sum up the characters, e.g. in an index, to get a proper comparison of the selections and the standards. In these experiments weight has been given to yield, fruit size, fruit quality, shelf life, storage ability and resistance to pear scab. From the means of these characters the following recommendations have been made:

'Moltke'

Accumulated yield per tree in the first seven years in the field was 26.6 kg, mean fruit size was 149 g, and mean date of harvest was 23 September. Fruit quality was best in October, and average fruit quality for this cultivar was regarded as acceptable in this month exclusively. Shelf life was medium in the months October to December. 'Moltke' was heavily damaged by bugs in one year at Ullensvang.

BP 8565: (Clapp's Favorite x Conference)

This selection performed differently at Njøs and Ullensvang as the yield at Ullensvang was considerably higher than that at Njøs. The mean accumulated yield per tree was 18.6 kg, and the fruit size was 124 g. Mean date of harvest was 7 October, which is rather late for Norwegian growing conditions. Fruits of BP 8565 scored high in fruit quality in October and December, but shelf life seem to be too short in December.

BP 9357: (Packham's Triumph x Herzogin Elsa)

The accumulated yield for this selection was 23.8 kg per tree and fruit size averaged 117 g. The date of maturity is very late as the mean date of harvest was 13 October. This is more or less a mean of fixed dates, as the latest harvest is dependent more on date and climate in the specific year than on evaluation of maturity. Fruit quality of BP 9357 was very good in October and December, shelf life was very good in October and November and acceptable in December. In spite of the late maturity this selection had acceptable fruit size and fruit quality. However, it is probably a selection for a warmer climate.

BP 10273: (Conference x Bonne Louise)

The merit of this selection is best characterized by its outstanding fruit quality in some judgements. Date of harvest was 16 September, i.e. one week before 'Moltke', which is very favourable for growers in Western Norway as far as harvest season is concerned.

Yield capacity was slightly lower than that of 'Moltke', with an average of 24.2 kg accumulated per tree, and fruit size was slightly bigger, averaging 159 g. The fruits are large and somewhat resembles the cultivar 'Conference'. Fruit quality was regarded as very good in October and December, but shelf life seemed to be too short in December.

BP 10104: (Herzogin Elsa x Conference)
Mean accumulated yield was 25.5 kg and fruit size 103 g for this selection. The average date of harvest was 27 September, and fruit quality was regarded as good in October and December. Shelf life was acceptable in December but not in January.

BP 9627: (Herzogin Elsa x Packham's Triumph)
This selection has a poor appearance, but is regarded as promising because of its outstanding shelf life and fruit quality after long term storage. The accumulated yield per tree was 16.6 kg and fruit size 130 g. Harvest date is late, the average picking date in these experiments was 9 October. Fruit quality is not acceptable when the fruits are not ripe, and proper ripening demands more than one week if fruits are marketed before January. BP 9627 is susceptible to pear scab, but it should be further tested because of its outstanding storage capacity.

Propagation material

A limited amount of virus-free material will be available for BP 8565, BP 9357, BP 9627 and BP 10273 in 1994, while BP 10104 will be delayed until 1996. Material may be obtained after signing an agreement with Balsgård.

SUMMARY

In two experiments at Ullensvang Research Station and substation Njøs, 46 selections from the Balsgård pear breeding programme were evaluated together with the standards 'Keiserinne' and 'Moltke'. The experiments were carried out in the years 1985 to 1991, and five promising selections were identified. Dates of first flower ranged from 20 April to 12 May and dates of harvest ranged from 28 August to 13 October. 'Moltke' had a higher accumulated yield than 'Keiserinne', and the highest accumulated yields were observed for the selections BP 895, BP 10529, BP 8881 and BP 9328. The largest fruits were found for the selections BP 10273, BP 8039 and 'Moltke'. The best fruit quality was classified each month, and high scores (>5.5) were noted for the selections BP 704, BP 895, BP 8565, BP 9357, BP 9627, BP 10104, BP 10201, BP 10273 and the standards 'Moltke' and 'Keiserinne'. The best results with respect to storage capacity and shelf life were from BP 6963, BP 9627, BP 9676 and BP 10104. For resistance to biotic and abiotic factors, it was found that the selections BP 9627, BP 10201 and BP 10529 were highly susceptible to pear scab *(Venturia pirina)*, and the selections BP 8039, BP 9066 and BP 10201 were highly susceptible to fruit cracking. The selections BP 8565, BP 9357, BP 9627, BP 10104 and BP 10273 are regarded as the most promising ones. Limited virus-free material will be available, but an agreement with Balsgård will be necessary in order to obtain propagation material.

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Assessment of silage additives using fresh or freeze-stored silage crops

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The aim of this study was to discover whether freeze-stored grass could be used as a silage crop in wintertime experiments. Ten laboratory silos, continuously monitored for pH, redox-potential, temperature, gas production and oxygen consumption, were used in eight experiments. Grass-clover mixtures were ensiled untreated, treated with 85% formic acid (3.0 cm^3 kg^{-1}), Foraform (4.0 cm^3 kg^{-1}), Kofa Plus (2.5 g kg^{-1}) and the inoculant Natuferm (106 CFU g^{-1}). Freezing and thawing the grass resulted in a lower number of viable microorganisms on the crop and a 38% reduction in the content of water soluble carbohydrates (WSC). Released plant juice after freezing increased the availability of fermentable nutrients, thus freeze-stored grass fermented in much the same way as the corresponding fresh material. It is concluded that freeze-stored grass does provide useful information as an experimental silage crop. Formic acid treatment significantly reduced pH and ammonia-N content of the silages ($p < 0.05$).

Key words: Additives, fermentation, freezing, frozen grass, silage

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A laboratory silo system for continuous monitoring of some silage fermentation parameters was set up in Norway in 1986. One of the initial objectives was to investigate whether this equipment could be used even during winter time. In general, using freeze-stored grass in periods when grass is not available can save time and money in the screening of different treatments before more expensive trials are carried out. Grass from crops ensiled in summer 1986 and 1987 was therefore frozen for repeated experiments. The most common silage additives in Norway were tested on both fresh and frozen/thawed grass to study their effects on silage fermentation and to discover whether there were any interactions between fresh or frozen/thawed herbage and silage additives.

MATERIALS AND METHODS

Crops for silage

The crops were harvested from fields with mixtures of timothy (*Phleum pratense*), meadow fescue (*Festuca pratensis*) and red clover (*Trifolium pratense*). Grass for the winter

experiments was deep frozen in 20 kg bags and thawed at 0-5°C for about seven days prior to ensiling. Just before ensiling the thawed material was spread on a polythene sheet to be mixed and to reach room temperature. At the time of ensiling, this material was about 5°C cooler than the fresh material in the summer. The ambient temperature (17-19°C) was reached a couple of hours post filling. This procedure was thought to cause minimum losses while thawing.

The composition of the fresh experimental crops is presented in Table 1. The herbage in experiments A1 and A2 was contaminated by soil from harvesting. A high clover content and harvesting at an early stage of development produced crops rich in crude protein in experiments A-C.

Table 1. The composition of the experimental crops

Experiment	A1 & A2		B1 & B2		C1 & C2		D1 & D2	
Ensilingtime	s	w	s	w	s	w	s	w
Cutting date	2/9		25/5		22/7		7/9	
Cut No	3		1		2		3	
Stage of growth	leafy		leafy		leafy		timothy headed	
Harvesting/ Chopping	Direct flail		Direct long		Wilted chop		Direct flail	
Dry matter g kg^{-1}	152		168		467		179	
Crude protein g kg^{-1} DM	295		292		268		88	
Sugar [1] g kg^{-1} FM	14		20		39		13	
Fructan g kg^{-1} FM	1		2		5		24	
Buffering capacity m equiv kg^{-1} DM	475		385		410		230	
Grass species % of DM								
Timothy	30		40		25		9	
Meadow Fescue	50		53		52		88	
Red Clover	15		6		10		0	

s=summer,fresh grass w=winter,frozen/thawed grass
[1] sucrose + glucose + fructose

Silos and experimental design

Using the ten silos in the system meant that continuous monitoring of pH, redox potential, temperature, gas production and oxygen consumption could be carried out. The equipment is described in detail by Selmer-Olsen & Müller (1989).

Approximately 1 kg dry matter (DM) was hand packed into each silo. Additives were diluted with 50 cm^3 water, spread on the grass in layers and mixed thoroughly before ensiling. To simulate slow filling, limited amounts of oxygen (6-12 l) were infused during the first 1-2 days. The limited O_2 infusion and the heat losses from the silos coused the temperature to rise only by as much as 8°C above the ambient temperature. All gas results were corrected for the spare volume in the silos and presented per kilogram of DM.

Fermentation data were recorded for 60 days before the silos were emptied for chemical analyses. Experiment A1, however, ended after 36 days.
Eight experiments were carried out based on four different crops. The experiments are identified by a common letter and number 1 or 2 for fresh and frozen/thawed herbage, respectively (A1, A2, B1, B2, C1, C2, D1, D2). Since experiments A1 and A2 were ended at different times, they were excluded from the comparison of silages from fresh and freeze-stored grass. In each experiment duplicate silos were filled for every treatment. The treatments were:

U = Untreated control
K = Kofa Plus (2.5 g kg^{-1} fresh matter (FM))
F = 85% formic acid (3.0 cm^3 kg^{-1} FM)
Ff= Foraform (4.0 cm^3 kg^{-1} FM)
N = Natuferm (approx. 10^6 viable bacteria g^{-1} FM)

Kofa Plus consisted of approximately 120 g kg^{-1} hexamethylen-tetramin, 220 g kg^{-1} sodium nitrite, 45 g kg^{-1} calcium formate and approx. 600 g kg^{-1} sodium chloride. Foraform consisted of 220 g kg^{-1} ammonia formate and 480 g kg^{-1} formic acid. The inoculant Natuferm consisted of Lactobacilli and Pediococci. A freeze-dried pellet of bacteria was dissolved in water, pre-fermented for 24 h at 20-25°C in a 2 l nutrient broth, diluted with 23 l of tap water and applied to the grass at 3.5 cm^3 kg^{-1} fresh crop.

Chemical and microbiological changes in the grass upon freezing and thawing were not systematically investigated in these eight experiments. This was therefore investigated with four different crops in a separate study.
Values for fresh grass were obtained by microbiological analysis of the fresh samples, whereas chemical analyses were carried out on frozen samples. New samplings for analyses were taken after freezing and thawing.

Microbiological analysis
The microbiological analysis was carried out at the Norwegian Food Research Institute (Matforsk). Appropriate serial dilutions were spread in agar and organisms were counted as colonies (CFU). Total viable aerobic organisms were enumerated in blood agar, lactic acid bacteria (LAB) in de Man Rogosa Sharpe agar, *Enterobacteriaceae* in Violet Red Bile Glucose Agar and Clostridia spores in thiosulphatenutrientbroth after 10 min heating at 75°C.

Chemical analysis
The entire contents of the 5 kg silos were frozen, then chopped, mixed and sampled for chemical analyses. DM was determined by oven drying at 67°C for 24 h, stabilizing to room humidity over night, milling and further drying at 100°C for 4 h. DM was corrected for losses of 80% of volatile fatty acids and 100% of ethanol. Total nitrogen (TN) and ammonia nitrogen were determined by the Kjeldahl method (Kjelltec Auto, Tecator). Organic acids and ethanol were analysed by HPLC with a Bio-rad column HPX-87H at 30°C and 60°C. Water soluble monosaccharides and disaccharides in the grass were analysed by HPLC using column HPX-87P from Bio-rad at 60°C. The samples were

diluted five times, macerated in a food processor for three minutes and filtered through 0.45 μm pore size before injection. Fructan was analysed as fructose at 30°C after hydrolysis in 0.5 N HCl at 100°C for ten minutes and neutralization by NaOH. The increase in fructose compared to the non-hydrolysed sample was then corrected for fructose from hydrolysed sucrose. Degassed distilled water was used as the eluent and flow rate was 0.6 ml min^{-1}. The sum of monosaccharides and disaccharides and fructan is expressed as water soluble carbohydrates (WSC). Buffering capacity was measured according to Playne & McDonald (1966). The redox potential refers to a saturated potassium chloride calomel electrode as the zero point and not to a hydrogen electrode scale as stated by Selmer-Olsen & Müller (1989). The latter gives approx. 250 mV higher values (Moisio, personal communication).

Statistical analysis

Least square means (LSMEANs) were estimated by analysis of variance using the General Linear Models (GLM) procedure (SAS 1987). The effect of freezing and thawing grass prior to ensiling was tested in a model as follows:

$$Y_{ijkl} = \mu + A_i + B_j + C_k + B_j{*}C_k + e_{ijkl}$$

where

Y_{ijkl} = lth observation, μ = overall mean,
A_i = main effect of crop, i=1,2,3
B_j = main effect of freezing/thawing, j=1,2
C_k = main effect of additive, k=1,2,3,4,5
$B_j{*}C_k$ = interaction between freezing/thawing and additives
e_{ijkl} = residual error

The main effects of silage additives were analysed in a model including main effect of experiment and interaction as fixed effects. Data from experiments with frozen grass were excluded when analysing the effect of additives.

Chemical and microbiological changes upon freezing and thawing were analysed as paired comparisons of relative values (t-test).

RESULTS

Changes in the grass upon freezing and thawing

The mean effect of freezing and thawing on the chemical and microbiological composition of four crops is shown in Table 2.

The content of WSC was reduced by 37.7% upon freezing and thawing. The numbers of aerobic bacteria, lactic acid bacteria and enterobacteria were reduced to less than half, but with considerable variation between samples. Samples with high counts of LAB and enterobacteria as fresh grass had higher percentage losses of bacteria than samples with low initial counts. The numbers of Clostridia were low in all samples (10-50 g^{-1}) and were not reduced with freezing and thawing.

Table 2. The effect of freezing and thawing on the chemical and microbiological composition of the grass. Paired comparison (n = 4)

	Fresh samples [1)]	Mean change after freezing and thawing	p
DM g kg^{-1}	182	- 3.3 %	NS
CP g kg^{-1} DM	117	+ 8.2 %	NS
Sugar g kg^{-1} FM	14.9	- 9.5 %	NS
Fructan g kg^{-1} FM	15.4	- 61.8 %	< 0.05
WSC g kg^{-1} FM	30.1	- 37.7 %	< 0.01
Buffering capacity m equiv kg^{-1} DM	324	- 4.7 %	< 0.1
Aerobic bacteria CFU g^{-1}	$3.76 * 10^7$	- 55 %	NS
LAB CFU g^{-1}	$1.85 * 10^6$	- 74 %	< 0.1
Enterobacteria CFU g^{-1}	$8.44 * 10^5$	- 51 %	< 0.1

[1)] Chemical analyses were carried out on deep-frozen samples

The effect of frozen/thawed grass versus fresh grass on silage fermentation

The main effects of frozen/thawed grass versus fresh grass on silage fermentation and interactions with silage additives are presented in Table 3. LSMEANs and interactions are calculated from three different crops (B, C and D) and five silage additives.

Table 3. Oxygen consumption, gas production and chemical composition of the silages from fresh and frozen/thawed herbage. LSMEANs from expts B1, C1, D1 and B2, C2, D2, respectively

	Fresh	Frozen/ thawed	SEM	Main effect	Interact. with add.
O_2 consumption, l kg^{-1} DM	17.7	16.2	0.225	***	
(available)	(18.9)	(18.2)			
Temperature rise, °C	4.9	4.8	0.22		
CO_2 production, l kg^{-1} DM					
0-3 days	24.8	21.2	0.81	**	
4-10 days	7.8	6.5	0.53		
11-60 days	10.4	9.4	0.48		
Total	42.9	37.0	0.62	***	
Total H_2 prod.,l kg^{-1} DM	0.03	0.41	0.067	***	
Corr.oven DM,g kg^{-1}	248	258	1.55	***	
Effluent, cm^3 kg^{-1} DM	2	68	7.3	***	
Silage pH	4.75	4.60	0.052	*	
Redox, mV	-247	-273	11.1		*
NH_3-N g kg^{-1} TN	90.3	90.2	4.74		
Lactic acid, g kg^{-1} FM	16.2	14.4	0.63		
Acetic acid, g kg^{-1} FM	6.7	6.9	0.30		
Butyric acid, g kg^{-1} FM	0.0	0.1	0.04		*
Propionic, g kg^{-1} FM	0.14	0.08	0.02		
Ethanol, g kg^{-1} FM	3.7	3.6	0.30		
Sugar, g kg^{-1} FM	5.1	8.3	0.57	***	

Level of significance: * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

SEM = standard error of LSMEANs. Interaction: freezing*additive

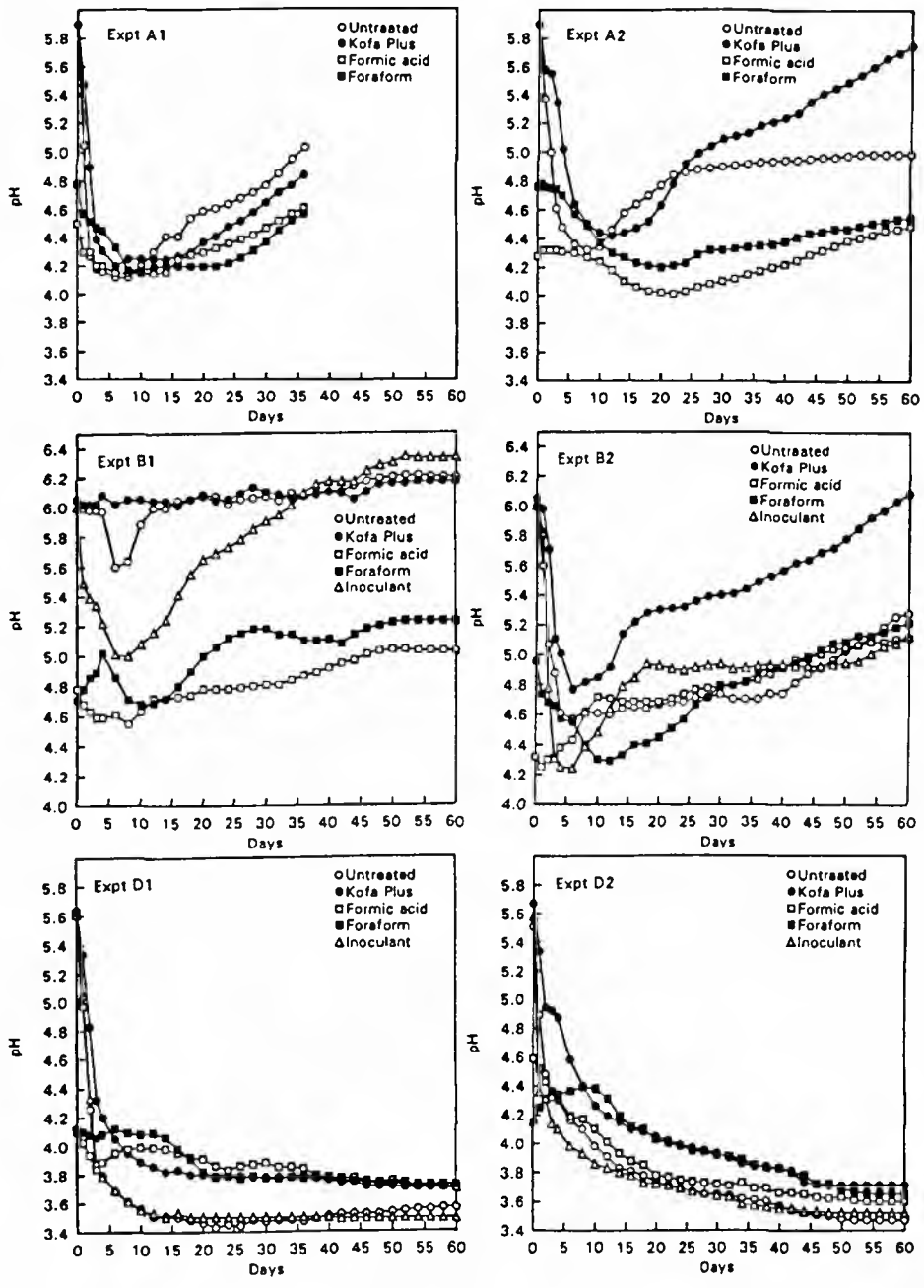

Fig. 1. Development of pH in the fresh herbage experiments A1, B1 and D1 and their respectively corresponding experiments A2, B2 and D2 with frozen/thawed herbage (n = 2)

The mean pH value was slightly lower for silage from frozen/thawed material than for silage from fresh crops. This was due to experiment B, when freezing obviously increased availability of substrate from the long grass. Development of silage pH over time for the fresh direct cut crops and the corresponding frozen/thawed material is shown in Fig. 1. Experiments C1 and C2 are not presented because the direct measurement of pH in the low moisture crops was impossible. The *in silo* pH-measurements were corrected to agree with the pH of the silage samples.

Plant juice released by freezing increased effluent losses from the silage and thus the silage DM content. In general the activity of the frozen/thawed herbage was reduced compared with that of the fresh herbage. This was reflected in the lower O_2 consumption and reduced CO_2 production during the first few days.

Other main effects of frozen/thawed versus fresh herbage were slightly more residual sugar in the silage and higher H_2 production. Significant interactions between effects of freezing/thawing the grass and silage additives were observed for the redox potential and butyric acid content. This was mainly due to the relatively poorer performance of Kofa Plus on the frozen/thawed herbage compared with the other treatments, which produced slightly better results. This was especially evident in experiment B with long grass (Fig. 1).

Butyric acid was found in experiments A1, A2, B1 and B2 only.

Then rise in H_2-production and pH was observed, whereas the redox potential was found to have dropped. The changes in these parameters indicated the initiation of butyric acid fermentation, which occurred after 16-18 days in experiment A1, 10-12 days in A2, 5-9 days in B1 and 20-30 days in experiment B2.

The effects of the silage additives on fermentation of fresh crops

The effects of the additives on O_2 consumption, rise in temperature, gas production and silage quality are presented in Table 4.

DISCUSSION

Changes in the grass upon freezing and thawing

Plant juice was released during the freezing and thawing processes and it was found that the grass was softer and wetter after freezing. The liquid was kept with the grass after thawing and the DM content did not change significantly. Compared with the fresh crop, the wetter conditions of the frozen/thawed grass should theoretically have been favourable to Clostridia, which have a lower limiting pH value when the water activity is high than when the water activity is low (Wieringa 1958). In the present study, however, much of the extracellular liquid was drained off as effluent after ensiling and the remaining surface moisture may have been commensurate with that for fresh herbage.

Frozen/thawed grass contained an average of 37.7% less WSC than the corresponding fresh grass. The content of monosaccharides was fairly constant, probably because hydrolysis of fructan continuously released monosaccharides, which subsequently were consumed in respiration. Thus the content of fructan decreased significantly. In general, Macrae et al.(1975) found only small changes in the carbohydrate content of the herbage during freezing and 24 h thawing. However, they found invertase activity while these

processes were going on. This made the levels of reducing sugar (monosaccharides) increase in contrast to the results from the present work. More extensive respiration should be expected from the longer thawing time in the latter. Macrae et al.(1975) found no changes in cell wall carbohydrates, *in vitro* digestibility or TN content. They found, however, that the proportion of soluble-N expressed as a percentage of TN decreased upon freezing and thawing. Their suggested explanation for this phenomenon was the breakdown of vacuolar membranes as a result of freezing. When mixing vacuolar and cytoplasmic contents a precipitation of proteins at the lower vacuolar pH may occur. In general, freezing *per se* does not damage enzymes; however, prolonged and repeated freezing may cause denaturation (Bøgh-Sørensen et al. 1986).

Table 4. Effects of additives on silage fermentation. LSMEANs from experiments A1, B1, C1 and D1

	U	N	K	Ff	F	SEM
O_2 consumption, l kg^{-1} DM (available 17.1)	16.5	16.8	16.3	16.3	15.6	0.31
Temperature rise, °C	4.2	3.8	4.1	4.2	3.5	0.41
CO_2 production, l kg^{-1} DM						
0-3 days	27.1a	27.4a	25.1a	22.4b	21.8b	0.88
4-10 days	5.2a	5.1a	6.9ab	8.9b	7.8ab	0.93
11-60 days	10.5	9.1	9.8	10.9	12.0	1.11
Total CO_2	42.8	41.6	41.8	42.2	41.7	1.75
Total H_2 prod.,l kg^{-1} DM	0.04	0.03	0.03	0.02	0.02	0.009
Corr.oven DM g kg^{-1}	222.3	219.2	228.2	227.5	230.6	3.21
Silage pH	4.94a	4.84abc	4.92ab	4.59bc	4.52c	0.125
Redox, mV	-299b	-233ab	-158a	-303b	-314b	32.4
NH_3-N g kg^{-1} TN [1]	104.5a	111.0a	97.3ab	72.1b	76.1b	8.90
Lactic acid, g kg^{-1} FM	15.0	18.0	14.2	14.3	13.6	1.37
Acetic acid, g kg^{-1} FM	8.0a	8.2a	7.2ab	5.9b	5.8b	0.61
Butyric acid, g kg^{-1} FM	0.5	0.3	0.2	0.2	0.2	0.17
Propionic acid, g kg^{-1} FM	0.2ab	0.3a	0.2ab	0.1b	0.0b	0.05
Ethanol, g kg^{-1} FM	2.4	2.1	3.4	3.8	3.4	0.54
Sugar, g kg^{-1} FM	4.7	3.8	5.0	4.7	6.1	0.52

U=untreated, N=Natuferm, K=Kofa Plus, Ff=Foraform, F=Formic acid

[1] Foraform and Kofa Plus are corrected for NH_3-N in the additive

a, b, c : LSMEANs not sharing common letters differ significantly ($p<0.05$). SEM = standard error of least square means

In the additional study on freezing and thawing, one effect was the reduced number of viable bacteria on the grass. According to Bøgh-Sørensen et al. (1986), rapid freezing, storage at about -20°C and slow cool (0-+5°C) thawing result in small losses of microorganisms compared with slow freezing, higher storage temperature and quick thawing. Spores survive more easily than viable bacteria.

In addition to the lower numbers after freezing, the lag phase of the microbial growth is extended (Bøgh-Sørensen et al. 1986). This is in agreement with the slightly slower initial fermentation in experiments A2 and D2 compared with that in experiments A1 and

D1, respectively (Fig. 1). A prolonged lag phase may be due to recovery of cytoplasmic membranes and DNA-nicks from the cold shock (Gray & Sørhaug 1983).

Silage from frozen/thawed grass versus fresh grass and interactions with silage additives
In general, silages made from frozen/thawed grass were similar to the fresh grass silages. This indicates that the increased availability of plant juice as a result of freezing to a large extent compensated for the loss of sugar in respiration during freezing and thawing.

Released plant juice from the freezing process ended up as effluent immediately after ensiling. Freezing the grass increased total effluent production and thus the DM content of the silage.

Reduced respiration appeared to occur in the frozen/thawed grass although lower O_2 consumption may have been a consequence of the slightly lower temperature during the first hours after ensiling.

Carbon dioxide production during the initial stages was lower for the frozen/thawed grass than that for the fresh grass. This difference was bigger than the difference in O_2 consumption, an indication that anaerobic processes were also delayed by freezing. From the lower total CO_2 production and higher residual sugar it appeared as though total fermentation was less extensive in the frozen/thawed grass than in the fresh material. It is likely that a difference in buffering capacity could explain this. The supporting study dealing with chemical and microbiological changes with freezing and thawing revealed slightly lower buffering capacity after thawing. In addition, increased effluent flow from the frozen/thawed grass may have removed buffering constituents, and the silages may have reached a stable pH with slightly less lactic acid and more residual sugar. This possible effect was clearly seen in experiments D1 and D2. An increase in residual sugar also occurred in experiments C1 and C2 with wilted grass. Then the frozen/thawed grass presented a purer lactic acid fermentation than the corresponding fresh grass.

A possible difference in rate of hydrolysis of hemicellulose during fermentation could have explained the higher content of residual sugar. Enzymatic hydrolysis of hemicellulose is more extensive at pH 6 than at pH 4 but is inhibited by low temperature (Dewar et al. 1963). There was no reason to believe that the freezing/thawing processes permitted extended hemicellulose degradation in the grass, since no increase in the content of arabinose or xylose was detected. Unfortunately, the silages were not analysed for hemicellulose.

For the purpose of using frozen/thawed grass as an experimental crop it is important to investigate the interactions between the effects of freezing the grass and the effects of the silage additives. The only parameters which revealed significant interactions were the butyric acid concentration and the redox potential. These two parameters are closely correlated since butyric acid fermentation produces H_2 and lowers the redox potential. The additive causing the interaction was Kofa Plus, which had a relatively poorer outcome on the frozen/thawed grass than on the fresh grass. All three chemical additives inhibit fermentation to some extent. However, the acid additives reduce pH directly, whereas Kofa Plus relies on fermentation. If freezing affected lactic acid bacteria more than Clostridia, the delayed drop in pH from the action of Kofa Plus may have allowed development of undesirable bacteria.

When the results from the three different crops were analysed separately, more

interactions were observed. The inoculated and the untreated long grass in experiment B ensiled relatively better after freezing (B2). This was probably due to increased availability of the nutrients for the bacteria after freezing. The improved lactic acid fermentation after freezing the grass probably delayed the butyric acid fermentation in experiment B2 compared with experiment B1. Long grass has previously been found to produce poor silage quality compared to chopped and minced grass (Ulvesli et al. 1965; Saue & Breirem 1969; Seale et al.1982). Experiments carried out with long grass may, however, yield information on how additives perform on round bale silage produced by a pickup baler.

One could expect the inoculant to compete better with the epiphytic microflora after freezing of the grass. This was not obvious, however. When the inoculant performed better on the frozen/ thawed herbage than on the fresh herbage it was also found that frozen/thawed herbage without additives ensiled better.

The effects of the silage additives

Well-fermented silage without additives was only produced from wilted grass (experiment E) and direct cut grass high in WSC and low in buffering capacity (experiment D). The relatively poorer performance of Kofa Plus on the frozen/thawed grass compared to the fresh grass would make the comparison of additives somewhat biased if all data were included. Therefore only data from fresh crop experiments are presented regarding the effects of silage additives.

In this study formic acid (85%) only tended to reduce respiration measured as O_2 consumption and rise in temperature. This is, however, a well-documented effect of formic acid (Henderson et al.1972; Nørgaard Pedersen & Witt 1973,1978; Pettersson 1988). This discrepancy may partly be explained by the availability of O_2, which in the present study was only 20-30% of what has been found to be consumed per kilogram organic matter during two days of free access (Nørgaard Pedersen & Witt 1978). Another explanation could be the spare volume in the silos, which served as a reservoir for the infused O_2. The O_2 was then gradually consumed also in the silages treated with formic acid. The O_2 availability was probably too small to cause a significant rise in temperature due to plant respiration. Heat loss from these small silos was rather high due to the large surface:content ratio. Heat was also lost via the silo lids, which were connected to the cooling system for condensation of water from the gas samples. Factors causing a minor rise in temperature in these silos would therefore cause a higher temperature on a practical scale.

During the first days of the experiments formic acid significantly reduced CO_2 production. Mo & Fyrileiv (1979) suggested that reduced CO_2 production with formic acid treatment could be due to inhibition of coliform bacteria (enterobacteria). In addition lactic acid bacteria are inhibited by formic acid treatment and thus CO_2 from heterolactic fermentation (McDonald et al. 1991). There was higher CO_2 production on days 4-10 from the formic acid treated silages than from the untreated and inoculated silages. This indicates that the fermentation was delayed and not totally restricted by formic acid (85%) at 3 l tonne^{-1}. Carbon dioxide production in the anaerobic phase of ensiling may be due to enterobacteria and Clostridia when pH is high, whereas heterofermentative lactic acid bacteria and yeasts may produce CO_2 also at low pH (McDonald et al. 1991). In experiment D, when the grass was rich in WSC, treatment with chemical additives restricted lactic acid fermentation whereas the content of ethanol in the silages increased.

Formic acid applied at a high rate (5 l tonne^{-1}) restricts fermentation and conserves sugar in the silage (Henderson et al. 1989). In the present study formic acid (85%) at 3 l tonne^{-1} only slightly restricted fermentation with direct cut grass; however, when combined with wilting the effect was stronger. Henderson & McDonald (1976) also found an enhanced preserving effect of formic acid with increasing DM levels.

Another significant interaction between the effect of silage additives and different experiments (crops) was found for pH.

With direct cut grass, which preserved poorly with no additive, formic acid-treatment resulted in the lowest silage pH. With easily fermented grass, however, chemical additives restricted fermentation and produced silages of higher pH than with inoculant or no additive.

Compared to the untreated control, the inoculant significantly increased the lactic acid content and reduced pH only in the wilted silage (experiment C). Consequently, this caused less residual sugar than with the other treatments. In the direct cut silages, the inoculant only tended to increase the lactic acid content. In the easily preserved direct cut silage the rapid fermentation with the inoculant saved sugar compared to the untreated control.

Formic acid and Foraform were the only additives that significantly reduced the content of acetic acid and NH_3-N as a percentage of TN. These are well-documented effects of formic acid treatment (McDonald et al. 1991).

In spite of the slightly slower acidification with Foraform than with 85% formic acid (Fig. 1) the silage composition was very similar. The main differences between Foraform and 85% formic acid were observed under good conditions (experiment D), where formic acid restricted fermentation slightly more than Foraform. This was reflected in a higher level of residual sugar with formic acid than with Foraform.

Kofa Plus did not improve fermentation quality in terms of the parameters measured in this series of experiments. It is something of a disadvantage that Kofa Plus is unable to aid acifidication either directly or indirectly.

In conclusion, experiments carried out with freeze-stored grass do give useful information on the effect of silage additives. However, experiments with frozen/thawed grass should not be entirely relied on, since there may be interactions between treatments and changes in the grass upon freezing and thawing.

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A note on the use of a short-duration serial digestibility study technique to assess changes in grass quality

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This study was carried out to examine the decline in grass quality associated with increasing plant maturity, using a continuous series of short-term digestibility trials. Six cuts of Italian ryegrass (*Lolium multiflorum* L.) were taken over a five-week period, starting at the three to four leaf stage and continuing until post-flowering. For the *in vivo* studies, three 18-month-old Dala rams were offered feed at approximately the maintenance level (39 g dry matter kg^{-1} $W^{0.75}$). Intake and faecal production were measured daily during the last seven days of each ten-day period. It was found that all digestibility coefficients decreased with increasing plant maturity. Dry matter (DM) and organic matter (OM) digestibility decreased by 11.3 and 13.8%, respectively, which was mainly attributable to the 16.3% decrease in crude (CF) digestibility. Net energy content decreased by 0.58 VEM kg^{-1} DM over the five weeks studied. The advantages and limitations of the technique are discussed.

Key words : Growth stage, Italian ryegrass, plant maturity

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Considerable time and facilities are utilized when conducting *in vivo* digestibility studies with ruminants. This is partly because the gut microflora has to become adapted to the feedstuff under examination and to ensure that sufficient time has elapsed during which accurate measurements of feed intake and faeces and urine production can be made. Such trials are generally conducted over 21 days and the data obtained over the last 7-10 days used to assess digestibility of the various feed components. However, when a series of similar feedstuffs (e.g. straw from different barley varieties) is being examined it is normally possible to reduce the introductory period, so that each complete study lasts about 14 days, without affecting the accuracy of the data obtained. In the study described here the well-documented decline in grass quality with increasing plant maturity was examined using a continuous series of digestibility trials, each lasting ten days.

MATERIALS AND METHODS

The study was conducted during 1990 using material harvested from a pure sward of Italian ryegrass (*Lolium multiflorum* L.) cv. Meritra, sown earlier in the year (5 April). Six tonnes of cattle slurry da^{-1} had been incorporated at ploughing and a further 7 kg N da^{-1} applied in mid-May. The grass was harvested from a plot situated centrally in a 1.0 ha field. Six cuts were taken at approximately weekly intervals, starting at the three to four leaf stage (12 June) and continuing until post-flowering (19 July). The material was packed in plastic sacks and frozen at -20°C until use. Each sack contained sufficient material to feed three sheep for one day. The growth stage, area harvested and fresh and dry matter (DM) yields obtained were recorded at each harvesting time.

The *in vivo* digestibility studies were conducted using three 18-month-old Dala rams (mean liveweight 55 kg). The test feeds were offered at approximately the maintenance level of feeding (39g DM kg^{-1} $W^{0.75}$) in the same order that they were harvested. The first study included a seven-day introductory period prior to the seven day feed intake and faecal collection phase. In all subsequent studies the introductory period was reduced to three days and the total study lasted 64 days. Urine was not collected. Feed allocations were prepared daily from material which had been thawed for at least 24 h. Samples were taken from each sack for DM determination. These were bulked and subsampled to obtain material for chemical analysis. Fresh weight of faeces produced was recorded and 10% taken for DM estimation. This material was then bulked and subsampled prior to analysis. Results from individual sheep were used to obtain the mean digestibility coefficients for each of the dietary components.

RESULTS

The harvesting scheme used allowed grass samples to be collected at various morphological stages from pre-emergence of the flowering head through to post-flowering (Table 1). The apparent decrease in DM content at harvesting was, on all occasions, due to rain. It was found that organic matter (OM) content, mainly crude fibre (CF), increased over the growing season, but nitrogen (N) and ether extract (EE) decreased (Table 2). The high initial levels of nitrogen are probably due to the fact that the first cut was taken only 24 days after fertilizing. Acid detergent fibre (ADF) levels were in the line with those of CF. All feeds were readily consumed and the sheep tolerated well their being held continuously in the metabolism crates for the duration of the experiment. The digestibility coefficients for all feed components examined decreased with increasing plant maturity. DM and OM digestibility decreased by 11.3 and 13.8%, respectively, which was mainly due to the 16.3% decrease in CF digestibility. ADF values decreased by 14.0%. Both DM and OM yield increased over the trial period but that of digestible OM increased at a declining rate (Fig. 1). Digestible energy content decreased with increasing maturity and when expressed in terms of net energy, calculated as VEM (Ekern et al, 1991), declined by 0.58 VEM kg^{-1} DM.

Table 1. Harvesting data

Harvest no.	Date	Yield (t/da)	Dry matter (%)	Growth stage
1	12.06	2.22	15.3	3-4 leaf stage, no heading
2	18.06	2.62	15.1	4-5 leaf stage, 5% heading
3	25.06	3.50	12.2	33% heading
4	3.07	4.21	13.2	67% heading
5	10.07	5.08	11.7	100% heading
6	19.07	4.92	16.1	post-flowering

Table 2. Chemical analysis[1]

Harvest no.	Organic matter	Ash	Nitrogen	EE[2]	Crude fibre	ADF[3]
1	0.875	0.125	0.046	0.051	0.196	0.206
2	0.882	0.118	0.039	0.053	0.210	0.218
3	0.883	0.117	0.033	0.043	0.240	0.251
4	0.893	0.107	0.030	0.031	0.254	0.262
5	0.903	0.097	0.025	0.033	0.302	0.316
6	0.912	0.088	0.024	0.024	0.289	0.306

[1] All values expressed as g g^{-1} dry matter
[2] EE - ether extract
[3] ADF - acid detergent fibre

Table 3. Digestibility coefficients[1]

Harvest no.	Dry matter	Organic matter	EE[2]	Nitrogen	Crude fibre	ADF[3]
1	0.831	0.869	0.663	0.858	0.930	0.914
2	0.828	0.855	0.649	0.819	0.924	0.927
3	0.781	0.825	0.554	0.792	0.888	0.884
4	0.760	0.797	0.492	0.771	0.847	0.843
5	0.771	0.799	0.562	0.744	0.859	0.859
6	0.735	0.753	0.473	0.713	0.778	0.786
SEM	0.017	0.018	0.034	0.021	0.023	0.021

[1] All values expressed as g g^{-1} dry matter
[2] EE - ether extract
[3] ADF - acid detergent fibre

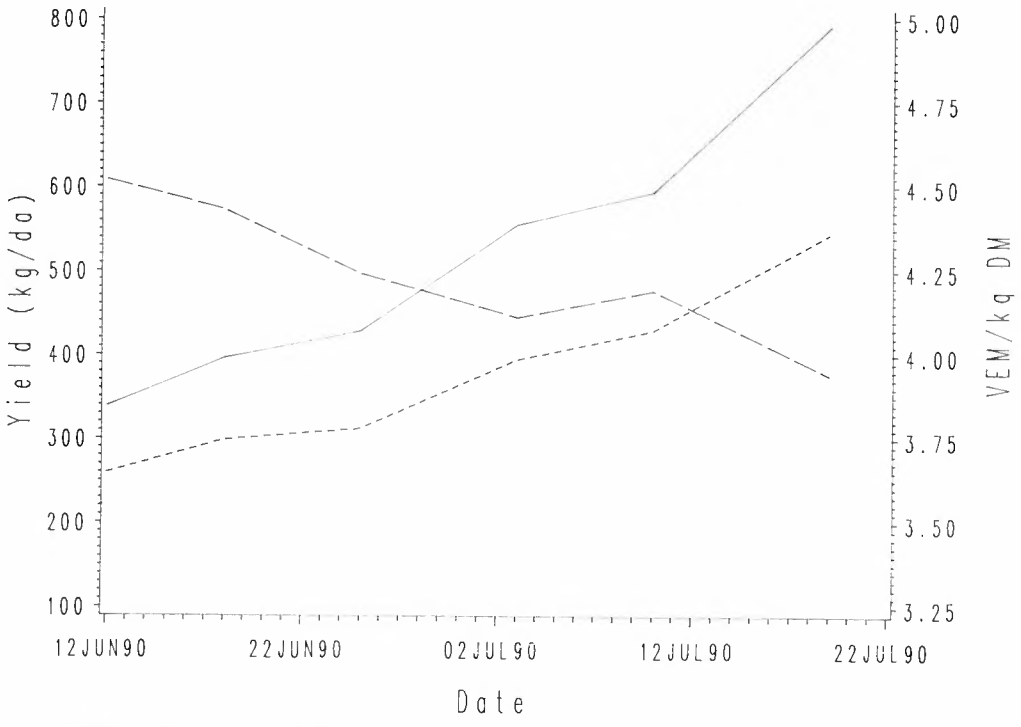

Fig. 1. Effect of harvesting date on dry matter (————————) and digestible organic matter (- - - - - - - -) yields (kg/da) and on net energy content expressed as VEM per kg dry matter (—— —— ——)

DISCUSSION

The *in vivo* technique employed allowed a more rapid assessment to be made of the change in grass quality over the growing season, than would normally have been possible. Even with 14-day periods the study would have taken an additional 20 days. An alternative method would have been to offer fresh grass, cut daily, with feed intake and faecal output measured continuously and the study divided into suitable periods for analysis. This, however, would have introduced considerable day-to-day variation in the quality of the grass offered. The use of weekly collection intervals, as in this study where sufficient material for each feeding period was obtained, avoided that problem. Where previous studies have investigated the effect of test period duration on digestibility (Blaxter et al 1956; Mandell et al 1987), these have examined the faeces collection period rather than the introductory phase. In this study the accepted standard collection period of seven days was used, while the introductory period was reduced. As there were only slight differences between consecutive feeds, only minor adaptations to the rumen microflora were required and therefore the use of a reduced introductory period did not appear to have adversely affected estimation of the digestibility coefficients. Using a shorter than normal period has the advantage that less feed material need be produced, stored and offered and that a greater number of studies can be conducted where facilities are restricted. However, it has

to be stressed that the feeds used in this study were very similar, of a high quality and readily consumed. Under circumstances where refusals are expected or where differences between feeds are considerable, then a longer test period is required. An extended period would also have to be utilized if minor differences between two similar feeds are to be detected.

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Effects of formic acid on performance in growing pigs

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Three experiments in which formic acid was added to the diet of pigs were carried out on 60 growing pigs. Formic acid (85%) was added to a commercial concentrate mixture in the amounts 0.6 and 1.2%, based on dry feed weight. Daily weight gain, feed conversion efficiency and meat quality were recorded. In Experiment 1 the addition of 0.6% formic acid to the diet in the period from 28 to 100 kg live weight resulted in an improvement in growth rate and feed conversion efficiency of about 8% compared to the control group on dry feed only ($p < .05$). The addition of 1.2% formic acid resulted in an (insignificant) improvement of about 2.5%. Experiment 2 started at weaning. No positive effect of adding 0.6% formic acid was observed from weaning to 20 kg live weight, but in the growth period from 20 to 100 kg live weight an improvement in daily gain and feed conversion of 11% ($p < .01$) was observed. In Experiment 3 an (insignificant) improvement of about 2.5% in daily gain and feed conversion was found. No effects on gastrointestinal diseases or on the meat quality were observed.

Key words: Feeding, formic acid, growing pigs, performance

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Feed costs account for the major part of the expenses in pig production (English et al. 1988). To get maximal revenue from the production, it is important to reduce the feed costs, to improve the growth rate and to improve the meat quality in the pigs.

A large number of feed additives have been used to improve growth rate and feed conversion efficiency in pigs, but many of these are hardly acceptable, whether from an ethical point of view, or because of the risk of pollution of the environment or because of residual concentrations in the meat (Weakland et al. 1988/89).

For quite some time the addition of acids has been used for preservation of feeds. More recently it has been shown that organic acids have a beneficial effect on health condition and growth rate in weaning pigs as well as in sows (Bolduan et al, 1990; Falkowski & Aherne 1984; Kirchgessner & Roth 1982; Eckel 1990; Kirchgessner & Roth 1988). This effect is not solely caused by a lowering of the pH in the feed, as it has been found that supplementation of the feed with phosphoric acid does not have the same beneficial effect (Roth & Kirchgessner 1989).

In growing pigs several feeding trials have been conducted to study the effect of

adding organic acids and the salts of organic acids to the feed (Kirchgessner & Roth 1988; Kirchgessner & Roth 1989). A positive effect has been observed in most of these experiments.

The present experiments were undertaken to study the effects of the addition of formic acid on performance in growing pigs fed on a commercial ration of concentrates.

MATERIAL AND METHODS

Animals

At Dal Research Farm, Norwegian College of Veterinary Medicine, three feeding experiments were carried out on 60 growing pigs. Experiment 1 started when the pigs had a live weight of about 28 kg; Experiment 2 started after weaning at five weeks (live weight 13 kg); and Experiment 3 started at a live weight of about 20 kg. All three experiments continued up to slaughter at around 100 kg live weight.

All the pigs used in the experiments were born on the farm. In Experiment 1 the animals were crossbred Norwegian Landrace x Yorkshire, in Experiments 2 and 3, Norwegian Landrace. The animals were confined indoors in pens with partly slatted floors. Sawdust was used for bedding.

From weaning to 20 kg live weight the pigs were fed in groups and after that the animals were placed in pens, six animals in each, equipped with crates for individual feeding of the animals.

The groups of pigs were as equal as possible with respect to average live weight, sex and parental origin and were allotted to experimental groups at random.

The pigs were weighed every second week.

All the animals were slaughtered at an abattoir at about 100 kg live weight. After slaughtering, the carcass weights and the meat percentages were recorded. Based on carcass weights, the live weights were corrected to a dressing percentage of 73. Daily weight gain and feed conversion efficiency were then calculated.

Feed

The animals were fed a commercial concentrate mixture (for the feed composition, see Table 1).

The animals were hand-fed twice daily according to a standard based on average live weight. The standard rate is almost that of appetite feeding. The feed was weighed out every second day for each pig and each feeding.

In addition to the water mixed with the concentrate for some of the groups, all animals had free access to fresh water from drinking nipples, one placed over the feeding trough and another placed near the dunging area.

Experimental design

A control group (1) was given dry concentrates without any supplementation. Another group (2) was given the same amount of dry concentrates, mixed with water (1 part dry meal to 3 parts of water) before feeding. In the experimental groups (3 and 4) the dry feed was mixed with the same amount of fluid as that in group 2 but with formic acid added.

The percentages of formic acid, based on the weight of dry feed, are given in Table 2.

The formic acid used in these experiments was designed for grass silage production (containing 85% formic acid, produced by NOFO A/S, 1430 Ås).

Experiment 1

The experiment started after an introductory period of 12 days, when the pigs were accustomed to the experimental feed.

Table 1. Composition of the commercial feed mixture used

Ingredients	Percent
Herring meal	0.60
Meat bonemeal	6.00
Fish silage	2.00
Soybean meal, extracted	2.63
Barley	40.80
Oats	40.00
Wheat bran	4.50
Molasses	1.00
Fat	1.50
Salt	0.25
Trace element premix*	0.10
Vitamin premix*	0.40
Amino acids**	0.22

* Supplying per kg feed: 50 mg Fe, 40 mg Mn, 70 mg Zn, 10 mg Cu, 0.5 mg I, 0.20 mg Se, 3.000 IU VIT A, 400 IU Vit D_3, 40 mg Vit E, 3 mg Vit B_2, 0.02 mg Vit B_{12}, 10 mg pantothenic acid.

** 0.16 % L-lysine and 0.06 % L-treonine.

Calculated contents:	
Energy, fattening units	94.0 per 100 kg feed
Crude protein	15.30%
Crude fat	5.30%
Calcium	0.70%
Phosphosrus	0.61%
Sodium	0.15%

Experiment 2

In this experiment the pigs were allotted in groups immediately after weaning. The animals were fed in groups until they reached about 20 kg live weight, when they were moved to the pens for individual feeding.

Table 2. Feeding of the groups in the experiments

	Group 1	Group 2	Group 3	Group 4
Experiment 1	Dry feed	Dry feed + water	Dry feed + 0.6% formic acid	Dry feed + 1.2% formic acid
Experiment 2	Dry feed	Dry feed + water	Dry feed + 0.6% formic acid	
Experiment 3	Dry feed	Dry feed + water	Dry feed + 0.6% formic acid	

Experiment 3
This was a replication of Experiment 1, without group 4, and with smaller pigs at start.

Statistical analyses
The groups were compared statistically by means of Student's t-test.

RESULTS

pH in the feed
In Experiment 1, pH in the feed in group 2 (mixed with water) was 5.5, in group 3 (0.6% formic acid) 4.7 and in group 4 (1.2% formic acid) 4.3.

In Experiments 2 and 3 the feed mixed with water had a pH of 5.6, and with the addition of 0.6 % formic acid, pH was 4.4.

Animal performance
The figures for initial live weight, daily weight gain, feed conversion efficiency and meat percentage are given in Table 3.

Experiment 1
At the start of the experiment, some of the animals in the 1.2% formic acid group were slightly reluctant to take the feed. This may have been due to uneven mixing of the water/formic acid mixture, but it did not occur later.

One pig in group 2 died suddenly after about one-and-a-half month into the experiment. Necropsy revealed a subaortal stenosis. One pig in group 4 was subject to tail-biting for a short period at the start of the experiment, but since this did not seem to have an influence on performance, the animal was included in the material.

The daily weight gain and feed conversion efficiency were significantly better in group 3 than in group 1 ($p < .05$) and in group 2 ($p < .001$). For the other figures analyzed, no significant differences were found.

Experiment 2
During the first weeks after weaning, when the animals were kept in groups without individual feeding, some of the animals in the groups fed the slurry (groups 2 and 3)

refused to eat part of the feed for some days. This caused a reduced growth in these groups compared to the group on dry feed during this period.

Table 3. Live weight, daily gain, feed conversion efficiency and meat percentage, average values ± standard deviation, and relative figures compared to Group 1

	Group 1	Group 2	Group 3	Group 4
Experiment 1				
Initial weight, kg	28.3±3.7	27.8±2.4	28.4±1.0	28.6±4.1
Relative figures	100	98.2	100.3	101.1
Daily gain to slaughter, grams	840±58[a]	816±35[c]	909±2[ac]	859±48
Relative figures	100	97.1	108.2	102.3
Feed conversion efficiency	2.69±0.19[a]	2.75±0.11[c]	2.47±0.03[ac]	2.59±0.15
Relative figures	100	102.2	91.8	96.3
Meat percentage	59.2±2.64	61.0±0.71	60.7±1.21	60.0±1.67
Relative figures	100	103.0	102.5	101.4
Experiment 2				
Live weight at weaning, kg	13.2±2.0	13.3±1.7	13.3±1.5	
Relative figures	100	100.8	100.8	
Daily gain, weaning to start of indvidual feeding, grams	443±42	386±19	381±116	
Relative figures	100	87.1	86.0	
Feed conversion, weaning to start of individual feeding	0.90	1.04	1.05	
Relative figures	100	115.6	116.7	
Live weight at start of individual feeding, kg	21.6±2.4	20.6±3.6	20.5±3.4	
Relative figures	100	95.4	94.9	
Daily gain, from start of individual feeding to slaughter, grams	800±30[b]	834±27[a]	892±58[ab]	
Relative figures	100	104.3	111.5	
Feed conversion	2.70±0.10[b]	2.59±0.08[a]	2.42±0.17[ab]	
Relative figures	100	95.9	89.6	
Meat percentage	59.5±1.38	59.0±1.79	61.5±2.17	
Relative figures	100	99.2	103.4	
Experiment 3				
Initial weight, kg	19.4±3.6	19.4±5.0	19.3±5.6	
Relative figures	100	100	99.5	
Daily gain, grams	795±51	788±55	818±56	
Relative figures	100	99.1	102.9	
Feed conversion	2.57±0.17	2.59±0.17	2.50±0.17	
Relative figures	100	100.8	97.3	
Meat percentage	60.7±2.16	59.8±1.72	60.3±0.82	
Relative figures	100	98.5	99.3	

Figures with identical letters in a row differ significantly, a) $P \leq 0.05$, b) < 0.01, c) $p < 0.001$

In group 2 tail-biting occurred before the animals were moved to the pens for individual feeding. The smallest pig in the group tended to be a tail-biter. This pig was removed from the group and was kept in a separate pen for the rest of the experiment, and by mistake it was not sent for slaughter along with the rest of the group. The live weight at slaughter for this animal is therefore calculated on the basis of live weight at the last weighing. The tail-biting did not seem to have a significant influence on the performance of the pigs.

Daily weight gain and feed conversion efficiency from 20 kg live weight up to slaughter were significantly better in group 3 than in groups 1 ($p<.01$) and 2 ($p=.05$). For the other figures analyzed, no significant differences were found.

Experiment 3

As can be seen from Table 3 there was a greater variation in live weight among the animals in this experiment than among those in the other experiments.

Also in this experiment there were some minor problems with tail-biting, but none of the animals displayed any clinical signs of illness as a result of this.

Group 3 had the highest daily weight gain and the best feed conversion efficiency, but no statistically significant differences were found.

DISCUSSION

The beneficial effects which were observed after the addition of organic acids to concentrate mixtures for pigs are considered to be due partly to a reduction in micro-organisms in the feed and hence in the stomach of the pigs (Morgenthum et al. 1989) and partly to an improvement in the activity of the digestive enzymes of the pig, mainly pepsin, thus increasing the digestibility of the feed (Burnell et al. 1988). Organic acids will also supply the feed in an energetic way as they will enter the citrate cycle (Kirchgessner & Roth 1988).

In the present experiment the addition of 0.6 or 1.2% formic acid caused a reduction in pH in the concentrate mixture from about 5.5 to 4.5 and 4.3 respectively. At this low level almost all bacteria and fungi will have been killed (Morgenthum et al. 1989).

Kamphues (1987, 1990) has shown how the acidity in the stomach of pigs is affected by feeding and by the addition of organic acids. Inclusion of 0.5% formic acid caused a small reduction in pH in the stomach, greatest in the pyloric region. Risley et al. (1992) found a decrease in the acidity in the stomach after the addition of 1.5% citric or fumaric acid, but this decrease was not statistically significant.

The buffering capacity of the food will have a strong influence on the pH in the stomach. An inclusion of mineral salts like calcium carbonate in the diet will cause a marked increase in the pH.

For an optimal digestion of proteins in the digestive tract, a conversion of the pepsinogen produced in the stomach to pepsin is necessary. A pH lower than 5.0 is required for this conversion. The pepsin produced has its pH optimum between 1.8 and 3.5 (Swenson 1984). The end products of pepsin activity also stimulate the secretion of pancreatic proteolytic enzymes (Cornelius 1988).

Newborn piglets have a low production of hydrochloric acid in the stomach and a pH near the neutral point (Swenson 1984). pH drops gradually up to weaning, when a rise in

pH occurs (Schnabel et al. 1982). Concentrates have a higher buffering capacity than sow's milk, this is considered at least partly to explain the increase in pH in the stomach after weaning.

For these age groups there is thus a reduced resistance to bacteria invading the stomach along with the feed, and there will also be a reduced pepsin activity. The positive effect of formic acid seen in pigs after weaning may be caused by these two factors (Kirchgessner & Roth 1982).

In Experiment 2 the addition of formic acid had no beneficial effect on the pigs from time of weaning up to 20 kg live weight. The feed intake was reduced compared with that of the group fed dry concentrates and was equal to that of the group fed on dry concentrates mixed with water.

In all three experiments it was observed that the addition of formic acid had a positive effect in the growing period from 20 to 100 kg live weight. This effect was significant in Experiments 1 and 2. In Experiment 1, the effects of adding 0.6 and 1.2% formic acid were compared, and it was found that 0.6% had a significant positive effect whereas the addition of 1.2% showed a non-significant positive effect. It could thus be concluded that the addition of 0.6% formic acid in the growing period from 20 to 100 kg live weight gives the best effect.

The positive effect seen in the present experiment might be due to an increased enzyme activity in the groups with added formic acid compared to the control groups and an increase in the digestibility of the nutrients in the feed.

Kirchgessner & Roth (1988) have shown that the addition of organic acids to the feed causes an increase in the digestibility of the nutrients, especially the proteins. In other experiments such an effect on digestibility has not been found (Giesting & Easter 1991).

The beneficial effects of adding organic acids are especially great in unthrifty, underweight pigs (Bolduan et al. 1990). One side effect of the addition of formic acid is a reported improvement in the general health condition of the pigs (Skjervheim 1991).

In the present experiment the hygiene as well as the management standards were high, with negligible gastrointestinal disease problems. This could indicate that under field conditions, in herds with a disease problem, an even greater positive effect might be expected.

CONCLUSION

The addition of formic acid to a commercial concentrate mixture for growing pigs at a level of 0.6% based on dry feed weight results in an increase in the daily weight gain and an improvement in the feed conversion efficiency from 2.5 to 11%.

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Methods of monitoring the effects of air pollution on forest and vegetation of eastern Finnmark, Norway

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A monitoring system has been developed in order to reveal the actual forest health situation in areas of north-eastern Norway affected by air pollution from Russian industry. Sampling is carried out on objectively chosen sample plots. Damage to vegetation and trees has been observed in the forests bordering Russia. Epiphytic lichens on birch stems are absent and show decreased coverage. Analyses of humus and plant tissues have shown elevated levels of nickel and copper in Norwegian forests located near the Russian border. The effects of pollution decrease with increasing distance from the emission sources, but it has been found that the critical level for lichen growth has been exceeded in parts of the area.

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Parts of north-eastern Norway and parts of the Russian Kola Peninsula are heavily polluted by air pollution from Russian industrial sites in Nikel and Zapoljarny (Fig. 1). The emissions from these sites are estimated (Sivertsen et al. 1992) to be 270,000 tons of sulphur dioxide (in 1989). In addition, significant amounts of some heavy metals are also emitted, mainly nickel and copper. The forest ecosystem is exposed to a variety of natural as well as anthropogenic factors that affect many of its processes, and result in specific conditions at different organizational levels in the ecosystem. The factors may be grouped into three categories:

Predisposing factors, which weaken the system;

Inciting factors, which result in more damage the more the system is predisposed;

Contributing factors, which may expand the incited damage.

An example is the predisposition of trees to air pollution, which stresses them into becoming susceptible to frost, which may occur as an inciting factor, causing direct frost injury into which canker fungi might enter as a contributing factor, thus increasing the initial frost damage. The process is generally described as the Decline Disease Spiral (Manion 1981).

In the Norwegian territory next to the Kola industrial sites, the air pollutants are thought to act mostly in a predisposing way, and to a lesser degree in a direct, inciting way. The average concentration of pollutants in the atmosphere is usually low, but

nevertheless constitutes a chronic stress upon the vegetation, predisposing trees and other plants to various injuries of climatic origin. Deposition of air pollutants, especially sulphates, may cause increased leaching of essential nutrients (base cations) from foliage and soil, resulting in visible deficiency symptoms and reduced growth. Degradation of soils by acidification and mobilization of toxic aluminium ions can occur. Continued deposition of heavy metals can cause problems for nutrient uptake in the roots, possibly by interfering with the mycorrhizae.

Fig. 1. From the area close to Nikel (1991)

Occasionally, however, episodes of high concentrations of certain air pollutants, in particular sulphur dioxide, cause direct injury to growing plant tissues, resulting in chlorosis or necrosis of exposed foliage (Aamlid 1993). Such injuries may affect growth, and more so after reoccurrences in subsequent years.

Deposition of pollutants may also affect animals feeding in the territory. Heavy metals are especially negative when deposited on or taken up in forage plants and accumulated in the food web of the ecosystem.

Severe ecological problems have existed over a long period. After the "perestroika" these problems were exposed through open cooperation between the two countries. A Joint Norwegian-Russian Commission on Environmental Cooperation was established. This commission has established expert groups for several environmental issues (e.g., air, water, human health, terrestrial ecosystems). The present description relates to a project established under the Expert Group on studies of Air Pollution Effects on Terrestrial Ecosystems. In addition to the described one, there are several projects run by the terrestrial group, concerning air pollution influence on animals or using remote sensing as a tool to investigate the influence on vegetation. These projects are closely linked to each other, and they receive funding from national agencies (Kismul et al. 1992).

For Norway, Sweden and Finland, it has been a task of high priority to negotiate reductions in the Russian emissions. However, the expence involved seems to be too high for the new Russian Republic to agree with the suggested requirements. This was clearly announced by the Russian Minister for the Environment, in Kirkenes in September 1992. It is therefore likely that large emissions from the industrial plants will continue for several years.

The objective of the present project is to evaluate the effects of air pollution on the

terrestrial forest ecosystem in the border areas by means of monitoring, to detect critical levels for plant growth in the area, and to evaluate the effect of decreased emissions as a result of industrial changes. This paper describes the methodology and its application.

MONITORING SYSTEM

In order to reveal the actual forest health situation, a monitoring system has been developed. Using the experience gained from the Norwegian monitoring programme for forest damage (Horntvedt et al. 1992), a programme for monitoring the effects of air pollution on forest ecosystems in these boreal forest regions was established in 1988 (OPS 1989). It was based on objectively chosen sample plots. A number of parametres thought to reflect the conditions of various levels in the terrestrial ecosystem were adopted or modified from other monitoring programmes, or were specifically developed for the present programme.

Sample plot grid network

A system of sample plots is established, representative of the area reflecting air pollution influence on the forest ecosystem. The sample plots are objectively chosen by laying a grid over the area (Fig. 2). The distance between each plot is 4 km. Each plot is circular, radius 25 m. If the chosen location for a plot lacks trees, mosses or lichens, the procedure is to try one of four new locations, 50 m east, south, west or north of the original location. However, many possible plots are still rejected because of lack of media (fell in wet bogs, water systems, etc.). At each plot an origin is defined, usually a tree, a stone or a rock (Fig. 3). The origin is marked with red paint, and serves as the basis for other measurements and subplots inside the plot.

A general description of each plot includes:

- Position (UTM)
- Altitude (above sea level)
- Topography
- Vegetation type
- Site class
- Slope
- Exposition
- Stand density

MEDIA AND PARAMETRES

The methodological philosophy of the project is partly based on a nutrient cycling model and partly on a food web model. However, the zoological elements are not a part of the present project. An overview of the main media and parametres are shown in Table 1.

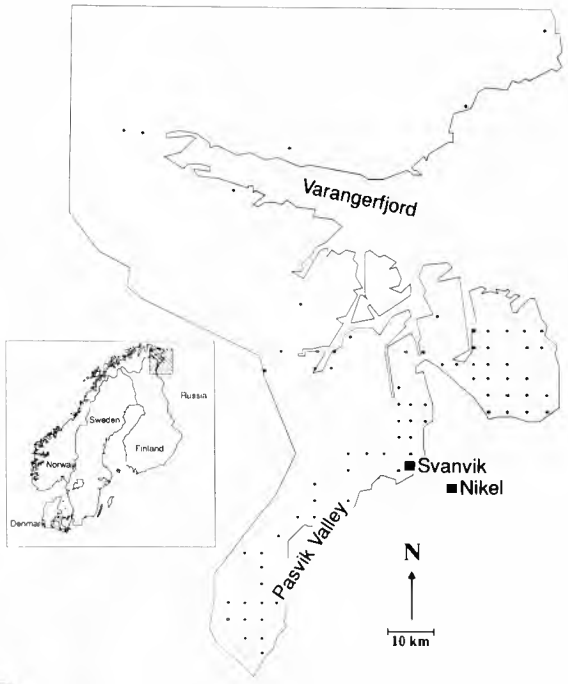


Fig. 2. The investigated area with sample plots

Fig. 3. From a sample plot

Table 1. Main media and parametres

Medium	Variable	Comments
Humus and mineral soil	Chemistry	Mineral soil at different depths
Field vegetation	Species	At permanent subplots 0.25 m^2 within the main plot
	Frequency	Occurence within the assessed square
	Injury	Symptoms due to several causes
Vegetation samples	Chemistry	Mineral nutrients and heavy metals
Epiphytic lichens	Species	At birch stems at four levels above ground
	Coverage	Assessed by use of "hit point" method
	Chemistry	Mineral nutrients and heavy metals
Trees	Species	The two main species, birch and pine
	Vitality	Crown density, crown colour, growth
	Injury	Symptoms due to several causes
	Chemistry	Leaves and/or needles, mineral nutrients and heavy metals
Deposition (incl. throughfall)	Amounts	At few selected locations, sampling each second week during vegetation period
	Chemistry	Mineral nutrients and heavy metals

SOIL SAMPLING AND ANALYSIS

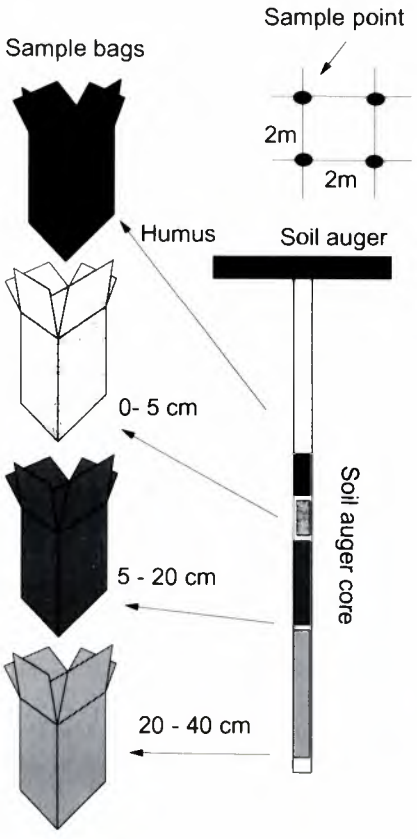
The soil sampling has to be representative of the plot. The method must also allow for later equivalent sampling, taken by independent persons. Because of limited resources the method must be easy to carry out. The chosen sampling procedure is based on these considerations.

The soil sampling is carried out in a subplot, 10 x 8 m, in a vegetation type representative of the plot. Sampling is done at each 2 x 2 m intersection (Fig. 4), giving 30 subsamples. Each subsample is divided into the following layers: humus horizon, and mineral soil from 0-5 cm, 5-20 cm and 20-40 cm according to the ECE recommendations (GEMS 1989). The sampled soil from each horizon or depth is mixed together before analysis, thus giving up to four soil samples from each plot. A representative soil sample is obtained from each layer at each plot in order to facilitate comparisons with later samplings, which are planned to be taken every fifth year.

The chemical analysis should reflect the pollution level and the mineral nutrition and the state of acidification in the soil as a medium for plant growth. The soil samples are therefore kept in dry and cool storage, until sent to the chemical laboratory as soon as possible.

Further pre-treatment for chemical analysis is performed according to Ogner et al. (1991). The humus and mineral soil are analysed for several elements. For mineral soil the following exchangeable cations and some other extractable elements are analysed in NH_4NO_3 extracts: Al, B, Ba, Be, C, Fe, K, Li, Mg, Mn, Na, P, S, Sc, Si, Sr and Zn. By using an ICP instrument, the following elements are analysed in the humus: Al, As, B, Ba, Be, C, Ca, Cd, Co, Cr, Cu, Fe, Ga, Ge, K, Li, Mg, Mn, Mo, Na, Ni, P, Pb, S, Sc, Se, Sn, Sr, Ti, V, Y and Zn. More details about the analysis methods can be found in Ogner et al. (1991).

Fig. 4. Soil sampling is carried out in a 2 x 2 m grid



VEGETATION SAMPLING AND ANALYSIS

Assessment of field vegetation

The assessment of the field vegetation should reflect the present situation concerning plant growth, vitality, diversity and influence by air pollution. Later, the analysis represents a platform for re-analysis with respect to the selected variables. It is therefore necessary to use a replicable method, irrespective of whoever carries it out.

The assessment of the field vegetation is based on permanent subplots inside the main plot. Each subplot is 0.5 x 0.5 m large. A total of 20 subplots is located into two rows, with a distance of 2 m between each subplot. The two rows are laid in a representative vegetation type for the plot. Each subplot is further divided into nine squares, 16.6 x 16.6 cm in area. When performing the assessment the subplot is defined using an aluminium frame (Fig. 5). Each subplot is permanently marked with two plastic sticks pushed down in opposite corners.

For each subplot the following general observations are made: plot number, subplot number, position according to origin, vegetation health, percent erosion, bare rock. This method is a slight modification of the methods used in other Norwegian monitoring programmes (Horntvedt et al. 1992; Fremstad 1992).

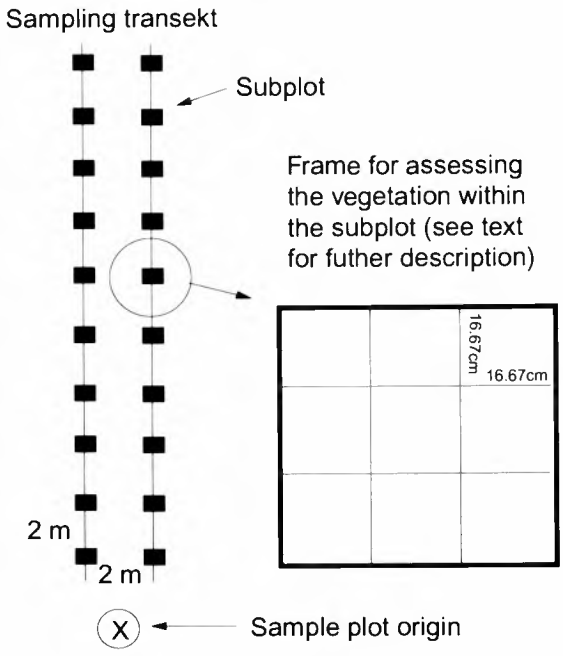


Fig. 5. Field vegetation is assessed in squares, 0.5 x 0.5 m in area

Within the subplot, observed species are noted, and in which of the nine squares they are found. Coverage or number of individuals of a species is not recorded, only the frequency within the square. The height of reindeer lichen (*Cladina* spp.) is measured inside each subplot (Fig. 5).

New analyses are planned every fifth year.

Coverage of epiphytic lichens

Epiphytic lichens are sensitive plants regarding air pollution. The sensitivity varies among different species. Lichen studies are therefore suitable for identifying air pollution levels within an area. Lichen studies are also used to identify air pollution that exceeds a certain critical level.

The analysis of the epiphytic lichen vegetation is carried out on four birch stems inside the plot (the four vitality trees of birch, see below). At each stem the lichen vegetation is analysed at four levels: 135 cm, 150 cm, 165 cm and 180 cm above ground level, done separately in the four main directions: north, west, east and south. The analysis is made by using a simple band with markers at each centimetre placed around the stem, and the number of markers hitting a single species is recorded for each aspect (Fig. 6). This method is also used by the Norwegian Monitoring Programme for Forest Damage (OPS 1988; Horntvedt et al. 1992). The percentage cover of each species on each trunk and level is then calculated from the measured circumference.

From trees other than those mentioned above, epiphytic lichens are sampled for chemical analysis to measure the level of pollutants.

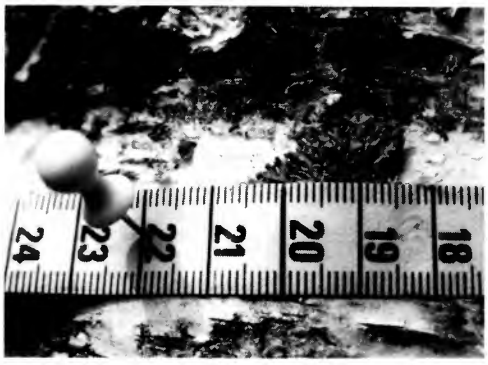


Fig. 6. Coverage of epiphytic lichens is assessed using a simple band around the stem. See text for details

A new analysis is planned every fifth year.

Vitality of trees

Trees are the most exposed receptors of air pollution in the area, and repeated damage associated with emissions of sulphur dioxide is reported. Such damage appears as necrotic leaf or needle tissue, and may lead to defoliation and reduced growth. Trees are also important as filters of polluted air.

The assessment of the vitality is made on selected trees. At each plot, four trees of birch and four trees of pine are objectively chosen by standing in the centre of the plot, facing north and selecting clockwise the first four predominant trees of each species. A selected tree should have a main stem. Each tree of each species is numbered (A, B, C, D) at breast height (1.3 m above ground level), the height and age is estimated, and diameter at breast height is measured.

The main variable is crown density, estimated in 1 % classes on the upper two-thirds of the living crown. The crown density of a tree is relative to a normal dense crown of trees in the region. (Fig. 7) (Aamlid et al. 1990; Aamlid et al. 1991.). This method of assessment, which is slightly modified in relation to the UN-ECE method used in the ECE countries (GEMS 1989; Aamlid et al. 1990), is used in Norway (NIJOS 1992; Solberg 1991) and Finland (Jukola-Sulonen et al. 1990). The most important improvement is the use of 1 % classes instead of 5 % classes. Crown assessment of birch is not described in the ECE manual. Only trained observers should carry out the assessment. Binoculars must be used for careful assessment. The trees must be inspected from all sides and at a distance of about one tree length. Obvious mechanical damage (snow break, wiping, shading or other associated effects) are disregarded. Crown colour is estimated by using the classes presented in Table 2.

Damage is recorded for each tree. Special damage recordings are, e.g., dead top and symptoms of sulphur dioxide (Fig. 8): brown needles (tips) or necrotic parts between the veins of leaves, as illustrated by Aamlid (1993). The assessments are made in late July or early August to avoid autumn colours on birch leaves.

Assessments of crown density, crown colour and damage are carried out annually.

Sampling for chemical analyses

A chemical analysis of vegetation is made to describe the level of pollutants absorbed or

adsorbed by plants, and to assess the nutrient condition of plants according to growth and vitality in relation to air pollution.

Table 2. Estimation of crown colour

Class	Percent of foliage yellowed	Description
1	0-10%	Normal colour
2	11-25%	Slight yellow/discoloured
3	26-60%	Yellow/Discoloured
4	> 60%	Strong yellow/discoloured

Fig. 7. Crown density is assessed on the upper two-thirds of the living crown

Generally, samples for chemical analysis are always handled using gloves made of polyethylene (changed for every sample), and stored in polyethylene bags or in approved paper bags until further treatment or analysis.

Vegetation samples are taken from the following species: *Betula pubescens*, *Pinus sylvestris* and *Vaccinium myrtillus*. With several years' frequency, the following species are sampled: *Hylocomium splendens* (or *Pleurozium schreberi*), *Cladina stellaris* (or other *Cladina* species), *Betula nana*. Other species are occasionally sampled: *Ledum palustre*, *Salix* spp., *Vaccinium uliginosum* and *Deschampsia flexuosa*. From mosses and lichens only the upper, living parts are sampled. From mosses, the last three annual shoots are analysed, while the upper half of the living parts from the lichens is analysed. From the other plant

species, only leaves or needles are analysed. From pine, the last three needle years are analysed separately.

Fig. 8. Brown leaves on birch caused by sulphur dioxide (Nikel 1991)

All plants are sampled from well-exposed individuals. As far as possible, all sampling is made inside the plot. However, if the vegetation cover is scarce, the sampling takes place close to the plot. Birch leaves and pine needles are sampled from trees near by the so-called "vitality trees".

Plant samples are milled to a required grain size according to the analysis (Ogner et al. 1991). Equipment used for preparation of samples is checked to avoid contamination. Milling equipment is carefully cleaned by use of ethanol.

The plant material is analysed for several chemical elements. To determine the total N, the Kjeldahl digestion is used. An ICP-AES instrument is used to determine the following elements (total) in plant material: Al, As, B, Ba, Be, Ca, Cd, Co, Cu, Fe, K, Mg, Mn, Mo, Na, Ni, P, Pb, S, Sc, Se, Sn, Sr, and Zn. More details about the analysis methods can be found in Ogner et al. (1991).

Vegetation samples for chemical analyses are taken annually.

DEPOSITION ESTIMATES

The deposition of sulphur and certain heavy metals is thought to be significant in parts of the investigated area. During the growing period of the year, precipitation is collected in

the investigated area.

At ten selected plots, north and west of the main pollution source, collectors are placed in open area, to collect *free-falling* precipitation (three collectors) and under birch trees to collect *through fall* precipitation (five collectors in the drip zone of tree crowns). The collectors are made of polyethylene (Fig. 9), and of similar type as those used in the Norwegian Monitoring Programme for Forest Damage (Horntvedt et al. 1992). However, here the collector is covered with aluminium foil to avoid heating since it is not placed down into the ground.

Throughfall and free-falling precipitation are analysed separately. The samples are analysed for pH, SO4-S, Al, B, C, Ca, Cu, Fe, K, Mg, Mn, Na, P, S, Si and Zn Analysis procedures are in accordance with Ogner et al. (1991). Results are compared with measured and modelled pollution levels (Sivertsen et al. 1992).

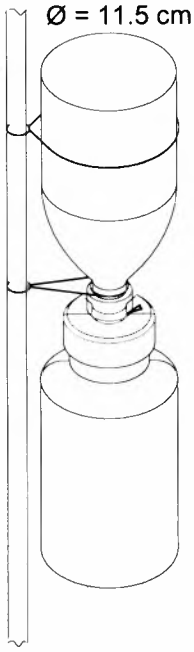


Fig. 9. Collector for precipitation (free-falling and through-fall)

The precipitation is collected continuously and sampled every second week.

EXPERIENCES AND COMMENTS

The establishment of the plot system started in 1988 (55 plots) and was later increased, so today there are 78 plots in the actual area (Fig. 2).

The density of plots is probably sufficient in the forested areas, but from a general point of view it would have been desirable to have more plots in the remote mountainous areas. However, these areas are not very accessible, and the cost of setting

up plots here would probably be very high. These plots would probably also be of less importance for the main questions, because these remote areas are less influenced by air pollution .

According to Kalabin et al. (1990) the annual deposition of SO4-S in the area surrounding the industrial site of Severonikel is about 5-7 g/m2. The deposition of metals (Ni+Co+Mn+Zn) is at a similar level. The SO4-S deposition at Svanvik (in Norway, 10 km west of Nikel) is approximately 0.3 g/m2 (SFT 1991). The results obtained in the present project reflect this situation.

Analyses of soil have shown that there are elevated levels of nickel and copper in the humus layer of Norwegian forests close to the Russian border (Fig. 10). Also, analyses of different plant species have shown elevated concentrations of nickel and copper in leaf tissue in these areas (Figs. 11 and 12). The epiphytic lichens on birch stems are absent in rather large areas in the eastern part of the investigated area (Fig. 13), and decreased coverage in even larger areas. Within the limited area investigated the forest vitality shows no clear improvement with increasing distance from the emission source in Nikel. On several occasions damage to vegetation and trees has been observed in the forests close to the Russian border. The damage has mainly been identified as partially brown leaves and needles (Aamlid 1992a; Aamlid 1993).

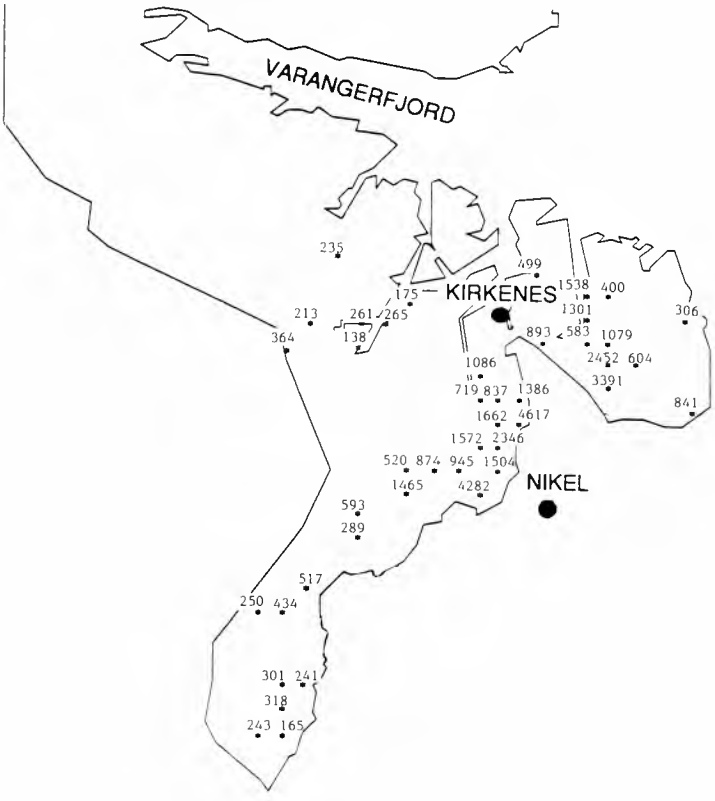


Fig. 10. Nickel (Ni) in humus (μmol/kg) 1989

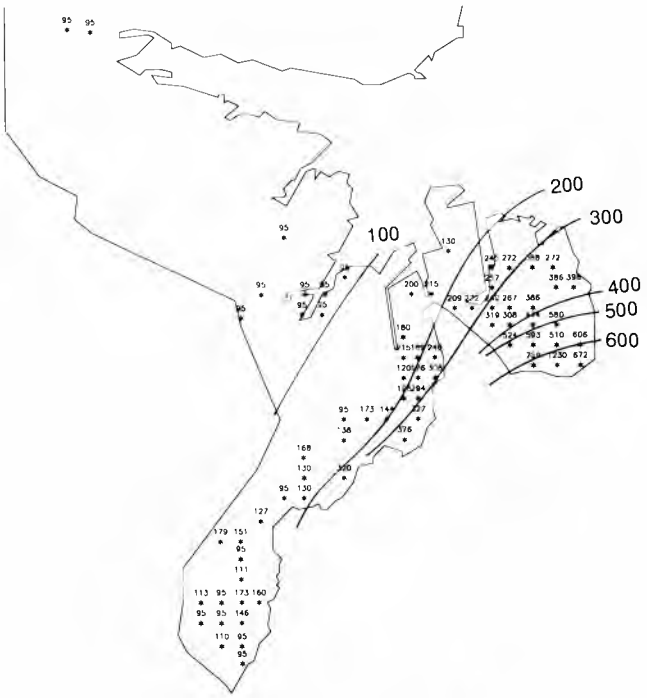


Fig. 11. Nickel (Ni) in leaves of *Betula pubescens* (μmol/kg) 1990 (from Aamlid 1992c)

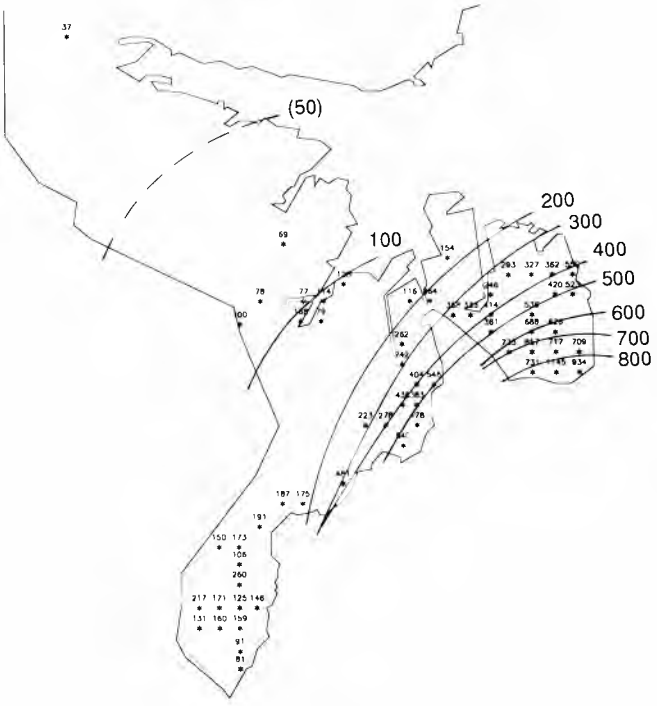


Fig. 12. Nickel (Ni) in leaves of *Betula nana* (μmol/kg) 1990 (from Aamlid 1992c)

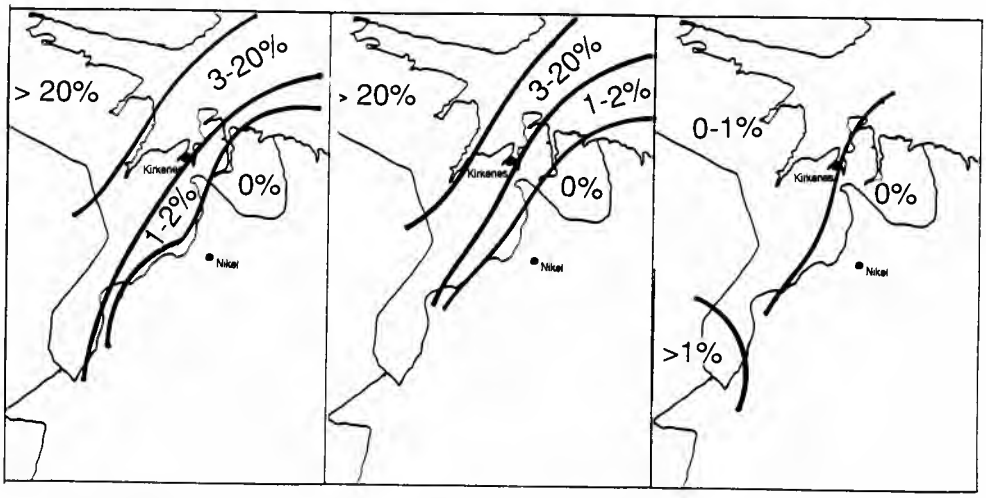


Fig. 13. Coverage of lichens (%) on birch trunks of *Hypogymnia physodes* and *Parmelia olivacea* (from Aamlid 1992b)

Results from the project are reported in more detail in different papers (Aamlid 1990, 1992a,b,c,d; Aamlid & Venn 1992).

In general, the results have shown a considerable influence of air pollution in the area, but the influence decreases with increasing distance from the emission sources. This is in accordance with modelled levels of air pollution in the area (Sivertsen et al. 1992). An example is what has been observed for the lichen flora. Critical levels of pollution for lichen growth are being exceeded in parts of the area. Chemical analyses of plant material have shown that uptake or accumulation of heavy metals and sulphur occur for all the analysed species. These results are also of importance in a food web context.

Continued monitoring will provide information on the state of the forest ecosystem, and will serve as a valuable basis for determination of critical levels in the area. The monitoring will also reveal the effect of changes in emissions, when they eventually occur.

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Effects of vegetation management on succession and on hardwood competition with Norway spruce (*Picea abies* L.)

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Compared with other relevant vegetation managements, overall foliage application with glyphosate caused the most pronounced reduction in number and coverage of the species involved. Regarding the field layer vegetation, however, the situation became, normalized within a 3-year period. The impact on treated vegetation appeared more pronounced after band- than after overall application of glyphosate. The invasion of seed-established *Betula* spp. and *Sorbus aucuparia* proceeded more slowly after clearcutting than after the overall application. Competition from suckers from the cut stumps had a major influence in this respect. The succession of the ecological important *Sorbus* species was stimulated by the cutting of *Betula* spp. combined with glyphosate treatment of the stumps. The overall chemical treatment caused the maximum growth reaction of Norway spruce, while clear-cutting resulted in the least reaction.

Key words: Clearcutting, foliage application, glyphosate, growth reaction, Norway spruce, stump treatment, vegetation succession.

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In Norway, where herbicides still play an important role in vegetation management in conifer plantations, glyphosate is the major herbicide used.

A research project initiated in 1983 by the Norwegian Institute of Forest Research, the Norwegian University of Agriculture and the Norwegian Plant Protection Institute, highlighted the influence of glyphosate application on vegetation succession, wildlife, water quality in streams and soil chemistry. The project came to an end in 1986 and the results were published in 1987 in the Scandinavian Journal of Forest Research, No. 2.

As many new questions arose from this project, the research into the ecological effects of glyphosate in forestry was continued in 1988 by the same research institutes involved. This new project ended in 1991. As was the previous project, this work was financed by the Agricultural Research Council of Norway.

This paper deals with foliage and stump application of glyphosate and alternative handcutting methods and their controlling effects on vegetation and succession of species.

The paper also covers the growth reactions of Norway spruce to the different vegetation managements used.

MATERIALS AND METHODS

Research areas

The research areas were situated at two locations in Hurdal (Kongliveien and Lundbyliveien) in the county of Akershus, at Ås in the same county and at Siljan in the county of Vestfold.

Kongliveien, Hurdal

The experimental area covered 1.5 ha, with a north-western aspect and an elevation of 400 to 420 metres. The site was of medium quality (site index G14) and the vegetation was characterized by *Betula* spp. and *Sorbus aucuparia* in the shrub/tree layer and *Deschampsia* flexuosa and *Vaccinium myrtillus* in the lower shrub/herb layer.

The area was clearcut in 1979 and planted with Norway spruce in 1981.

The experiment covered five experimental sites according to the following plan:

a. Foliage application with glyphosate (1 kg a.i./ha) from a helicopter in August 1988
b. Clearcutting of all hardwoods higher than 10-15 cm in August 1988
c. Clearcutting of all competitive *Betula* spp. and just a minor portion of *Sorbus aucuparia* with a subsequent stump surface treatment with 7.2% glyphosate
d. As for c, but without stump surface treatment
e. No vegetation management

Each year, after the treatments, in August-September, the following parameters were evaluated:

Hardwoods: The number, frequency and mean height of remaining trees, the initial number of stumps, annual number of suckers and mean height of suckers at the end of the experimental period. The frequency was defined as the percentage of the total number of plots within an experimental site having a given species.

Norway spruce: Number and height of the four highest plants per plot.

Shrub/herbs: Percentage plant coverage of the investigated species and the frequency, as explained under "Hardwoods".

The evaluations were based on 2 x 2 m plots at 4 m intervals centred on lines 6 m apart. The mean number of plots per experimental site was 100.

Lundbyliveien, Hurdal

The experimental area covered 3.2 ha divided into five experimental sites (40 x 200 m). The elevation was 350-420 m and of a north-eastern aspect. The site quality was high (G20) and the vegetation was characterized by a few *Betula* spp. in the tree layer and *Deschampsia flexuosa*, *Rubus idaeus* and *Vaccinium myrtillus* in the lower layer.

The area was clearcut in 1986/87 and planted with Norway spruce in 1989/90.

The main purpose of the experiment was to evaluate the reinvasion of vegetation after an overall application from a helicopter compared to band application from a hand-operated

knapsack sprayer with 1 kg a.i. glyphosate/sprayed per ha. A second question was focused on the effects of overall application two and three years after logging, respectively, with planting of Norway spruce the year after each of the applications. However, because of poor plant quality, 60-70 % of the plants died during the first few weeks after planting in spring 1989. Consequently, this part of the investigation was excluded, and the final plan covered the following treatments:

1. Overall application with 1 kg a.i./ha glyphosate in August 1988
2. Band application (1 m width of the band) in August 1988
3. No vegetation management

The evaluation of results was carried out as described for Kongliveien.

The size of the evaluation plots in this experiment was 0.5 x 2 m, centred on lines 5 m apart, giving 200 plots on each experimental site.

Aschjem, Ås

An area of 1.5 ha was covered with the same kinds of experimental plots as those described for Kongliveien, Hurdal, except for plot d. The elevation of the area was about 50 m and with a south-eastern aspect. The site productivity was quite high (G20). The area was logged in 1984 and planted with Norway spruce in 1986.

The predominant vegetation comprised a wide range of hardwoods such as *Betula* spp., *Sorbus aucuparia*, *Salix* spp., etc. The most characteristic species on the shrub level were *Galeopsis tetrahit*, *Pteridium aquilinum*, *Rubus idaeus* and various grasses.

An evaluation was carried out as explained for Kongliveien. The experimental basis for the evaluation composed 2 x 2 m plots centred on lines 6 m apart. The total number of plots within the individual experimental site was 102.

Siljan, Vestfold

The experimental area was 1.3 ha of moderate productivity (G14). The area was logged in 1980 and planted with Norway spruce in 1981. The site had an elevation of 200-230 m and was of a north-western aspect.

The dominant vegetation included hardwoods such as *Betula* spp. and *Sorbus aucuparia*, with *Deschampsia flexuosa* and *Vaccinium myrtillus* in the shrub layer.

The purpose and experimental plans were identical to those at Kongliveien, Hurdal. The number of plots per experimental site was 100.

The glyphosate used in these experiments was formulated as Roundup (360 g a.i./L).

RESULTS

Effects of vegetation management methods

Hardwoods

Kongliveien: The overall application of glyphosate (a) in 1988 significantly reduced the number of *Betula* seedlings by about 98% compared with that in the untreated site (Table 1). The frequency of plots with seedlings was also considerably reduced. In 1991 the number of *Betula*

seedlings had increased to about 1400 per hectare, about 4% of the number on the untreated site. The frequency of plots with plants practically doubled within the same period.

Table 1. Effects of various vegetation management methods on hardwoods. (a: Overall glyphosate application 1988, b: Clearcutting 1988, c: Selective handcutting combined with stump treatment, d: Selective handcutting and e: No vegetation management). Location: Kongliveien, Hurdal

Management	Year	Uncut, live trees > 10cm			Cut trees				*LSD (year)	
		No./ha	Frequency (%)	Mean height (cm)	No./ha			Mean height (cm)	No.	Mean height (trees)
					Stumps /ha.	Suckers /ha.	Suckers /stump			
					Betula spp.					
a	1989	525	12	96						
	1990	850	23	91						
	1991	1400	29	96					3033	2.54
b	1989	1935	33	54	6024	29735	4.9			
	1990	6042	60	36		25789	4.3			
	1991	6095	48	45		21530	3.6	45	3263	1.78
c	1989	1995	39	84	8266	13624	1.6			
	1990	3395	53	77		12788	1.7			
	1991	4151	65	94		12220	1.5	38	2710	1.54
d	1989	4673	73	91	15334	66283	4.3			
	1990	7857	85	92		50673	3.3			
	1991	7113	68	118		46250	3.0	42	3062	1.40
e	1989	27165	94	128						
	1990	30593	95	144						
	1991	32912	93	170					3036	1.19
*LSD (Management)	1989	2748		1.69						
	1990	3036		1.47						
	1991	3278		1.91						
					Sorbus aucuparia					
a	1989	100	3	60						
	1990	1025	18	56						
	1991	850	16	59					3062	1.98
b	1989	387	8	50	10512	26354	2.5			
	1990	1191	23	48		56298	5.4			
	1991	923	14	51		49550	4.7	19	3341	1.66
c	1989	7661	58	72	1248	1812	1.5			
	1990	12248	60	61		3534	2.8			
	1991	13257	64	57		4470	3.6	15	2826	0.72
d	1989	4494	33	75	397	543	1.4			
	1990	5149	36	68		896	2.3			
	1991	6518	33	65		800	2.0	23	3341	1.04
e	1989	6057	47	67						
	1990	8196	47	61						
	1991	7526	46	67					3109	0.85
*LSD (Management)	1989	2196		1.47						
	1990	3177		1.05						
	1991	4034		1.22						

*LSD ($P \leq 0.05$) value for comparison of means.

Clearcutting of all hardwoods (b) also significantly reduced the number of *Betula* seedlings the year after the treatment. The recovery after this treatment was somewhat faster than that after the glyphosate application. The treatment gave an approximately 50% lower mean height of seedlings and a more rapid increase in number from 1989 to 1990 compared to cutting of just competitive species (d) (Table 1). The number of suckers per stump was also higher for treatment b.

Stump treatment with glyphosate (c) caused about 50% fewer suckers per stump than treatments b and d (without stump treatment) and a similar increase in the number of seedlings from 1989 to 1991. On the other hand, the stump treatment caused a significantly higher mean height of seedlings than treatment b, probably because of reduced sucker competition.

Treatments b, c and d all showed a decline in number of suckers from 1989 to 1991.

For *Sorbus aucuparia*, Table 1 presents a situation opposite to that explained for *Betula* regarding the number of suckers per stump over time. A stimulating effect of stump treatment on the increase of *Sorbus* seedlings from 1989 to 1991, compared to treatment d is also presented in Table 1.

The results indicate that *Sorbus* is rather sensitive to competition from *Betula* spp. Cut surface treatment of *Sorbus* stumps had less effect than the treatment of *Betula*.

Overall application with glyphosate caused a faster re-establishment of *Sorbus* seedlings than that of *Betula* seedlings.

Siljan: The results as presented in Table 2 show tendencies regarding *Betula* spp. similar to those explained for the experiment at Kongliveien. However, clearcutting resulted in a greater reduction in the number of seedlings than that with the glyphosate application. The speed of reinvasion of new *Betula* seedlings from 1989 to 1991 was also significantly lower after the clearcutting. The frequency of plots with seedlings increased dramatically after both treatments, reaching the level of untreated plots by the third year. However, the mean height of seedlings after the two treatments was only about half that of seedlings in untreated plots. Glyphosate treatment of the *Betula* stumps showed less effect than in the Kongliveien experiment. The lower mean height of both seedlings and suckers after treatment d compared to treatment c (without and with stump treatment) may be a consequence of greater competition on plot d.

Sorbus showed a similar reaction to glyphosate application and clearcutting to that of the *Betula* seedlings, and a surprising decline in the number of seedlings from 1989 to 1991. Stump treatment had evidently better effect against *Sorbus* suckers in this experiment than at Kongliveien.

Ås: The results in Table 3 indicate that practically 100% of the *Betula* seedlings were killed the year after glyphosate application, with a rather sparse regeneration. The number of suckers per stump after clearcutting decreased drastically in the period 1989-91. This was also recorded after selective handcutting combined with stump treatment. This treatment had therefore only a small effect. All the vegetation managements in the Ås experiment significantly lowered the mean height of *Betula* seedlings within the experimental period.

The foliage application with glyphosate significantly reduced the number of *Sorbus* seedlings throughout the experimental period. According to the results recorded in Table 3, this was also true for the clearcutting treatment, with no significant difference between the two measures. Selective handcutting of *Betula* and competing *Sorbus* and with subsequent stump treatment, highly stimulated the growth conditions for the seedlings of this species in spite of the fact that regrowth from cut *Sorbus* stumps was reduced by 30-40% compared to site b.

Table 2. Effects of various vegetation control measures on hardwoods (see Table 1). Location: Siljan

Management	Year	Uncut, live trees > 10cm			Cut trees				*LSD (year)	
		No./ha	Frequency (%)	Mean height (cm)	No./ha Stumps /ha.	No./ha Suckers /ha.	Suckers /stump	Mean height (cm)	No.	Mean height (trees)
					Betula spp.					
a	1989	1800	20	79						
	1990	4600	29	56						
	1991	9950	100	38					3614	3.07
b	1989	775	17	69	8176	40215	4.9			
	1990	1100	21	78		32548	3.9			
	1991	950	100	106		28860	3.5	55	3614	4.10
c	1989	3900	63	116	8386	37136	3.9			
	1990	3625	60	149		33208	3.8			
	1991	3675	100	184		26750	3.2	64	3614	2.23
d	1989	8525	88	108	24538	106008	4.3			
	1990	8675	85	129		99387	4.1			
	1991	7875	100	158		84250	3.4	54	3614	1.89
e	1989	10625	51	157						
	1990	9850	53	185						
	1991	11000	100	196					3614	2.46
*LSD (Management)	1989	3042		2.38						
	1990	3565		2.80						
	1991	4236		3.05						
					Sorbus aucuparia					
a	1989	1325	18	48						
	1990	1150	15	41						
	1991	850	100	33					5265	1.92
b	1989	1075	19	75	9887	25035	2.5			
	1990	600	16	96		28146	2.8			
	1991	625	100	90		20810	2.1	42	5275	1.83
c	1989	16450	67	67	734	786	1.1			
	1990	19350	65	64		2235	3.1			
	1991	20850	100	60		580	0.8	38	5256	0.90
d	1989	12050	59	64	653	2087	3.2			
	1990	11250	56	58		3508	5.4			
	1991	13525	100	52		3300	5.1	27	5265	0.96
e	1989	22025	77	89						
	1990	25700	82	85						
	1991	30200	100	77					5265	0.82
*LSD (Management)	1989	4795		1.14						
	1990	5196		1.30						
	1991	5805		1.41						

*LSD ($P \leq 0.05$) value for comparison of means.

Table 3. Effects of various vegetation control measures on hardwoods (see Table 1). Location: Ås

Management	Year	Uncut, live trees > 10cm			Cut trees				*LSD (year)	
		No./ha	Frequency (%)	Mean height (cm)	No./ha Stumps /ha.	No./ha Suckers /ha.	No./ha Suckers /stump	Mean height (cm)	No.	Mean height (trees)
					Betula spp.					
a	1989		1							
	1990	29	1	30						
	1991	29	1	10						
b	1989	494	10	153	5125	31356	6.1			
	1990	581	17	183		14912	2.9			
	1991	378	9	262		11920	2.3	165	818	9.17
c	1989	646	12	165	6346	5208	8.2			
	1990	787	16	197		2287	3.6			
	1991	871	13	238		1830	2.9	148	804	8.38
e	1989	648	14	238						
	1990	2006	19	207						
	1991	2220	4	415					910	10.67
*LSD (Management)	1989	731		8.35						
	1990	1153		13.67						
	1991	629		17.09						
					Sorbus aucuparia					
a	1989	174	3	113						
	1990	1221	23	75						
	1991	1541	24	73					3709	4.80
b	1989	2035	36	111	14645	63834	4.4			
	1990	2006	41	115		65361	4.5			
	1991	1424	8	111		53830	3.7	108	3709	3.79
c	1989	16376	89	165	8708	20054	2.3			
	1990	18146	88	176		31653	3.6			
	1991	16910	73	193		23880	2,7	105	3646	1.78
e	1989	22253	91	204						
	1990	25803	94	214						
	1991	27722	95	270					4240	2.03
*LSD (Management)	1989	3457		2.99						
	1990	4126		2.55						
	1991	3895		3.74						

Other species registered on the experimental area at Ås, such as *Quercus* spp., *Prunus* spp. and *Sambucus racemosa*, were evidently easier to control by clearcutting than by glyphosate application.

Growth reactions of Norway spruce

Kongliveien

Height growth: From 1988 to 1991 the plant heights increased by 72%, 19%, 41%, 38% and 36% after the a, b, c, d and e treatments, respectively (Fig. 1).

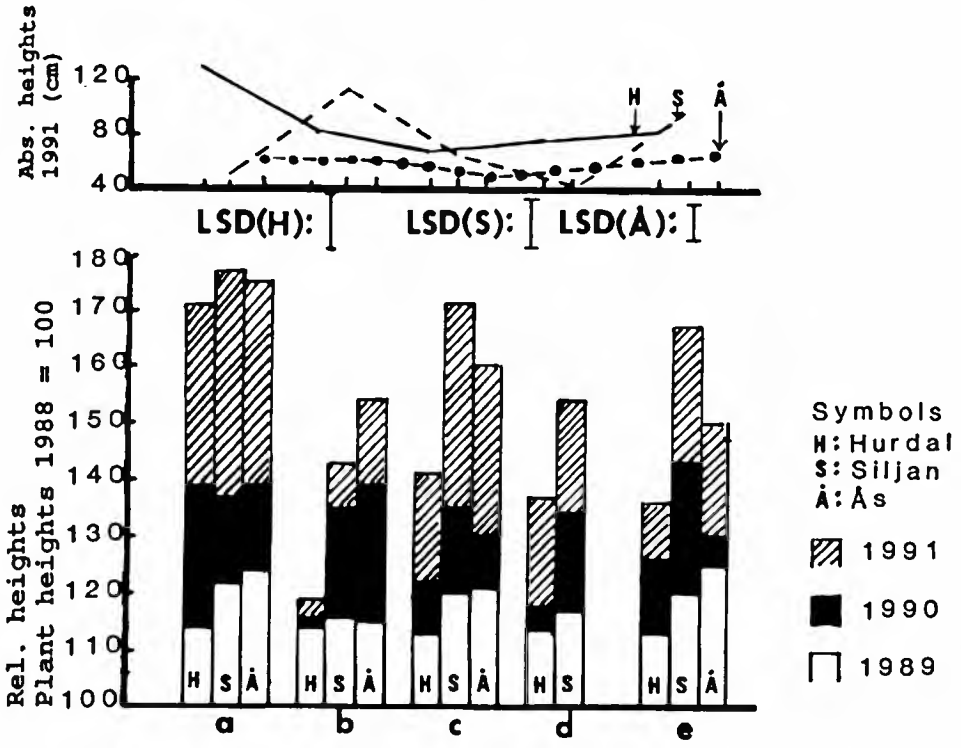

Fig. 1. Effects of various vegetation managements on the growth of Norway spruce (*Picea abies L.*). Upper figure: Plant heights in 1991; lower figure: Relative, yearly growth. Treatments in 1988: a) Overall application of glyphosate; b) Clearcutting of all hardwoods; c) Clearcutting of competitive hardwoods combined with stump treatment with glyphosate; d) As for c, without cut stump treatment; e) No vegetation management. Vertical bars represent LSD ($P \leq 0.05$) values regarding relative height levels in 1991 for comparison of treatments within a location

The overall glyphosate treatment gave significantly better results in 1991 than the other treatments and clearcutting had a significantly poorer effect than the other handcut methods and untreated plots.

Number: The number of spruce plants shows only a negligible increase after the glyphosate application within the experimental period 1989-91 (Fig. 2). On the other experimental sites, clearcutting of the hardwoods (b) caused a 31% increase, the selective handcutting combined with stump treatment (c) a 52% increase, the same treatment without stump treatment a 24% increase and the site with no vegetation management a 9% increase. Only the 52% increase appeared significant. Natural regeneration can be the only reason for the increase. Therefore it may be supposed that the glyphosate application had a depressive phytotoxic effect and the treatment with no vegetation management had a depressive competitive effect on the establishment of natural spruce seedlings.

Siljan

Height growth: The relative increase in plant heights from 1988 to 1991 was most pronounced after overall glyphosate application (79%) and selective handcutting with stump treatment (71%). But even on the untreated site, the corresponding increase was as high as 68%, indicating no strong correlation between the vegetation managements and height growth reactions within the experimental period.

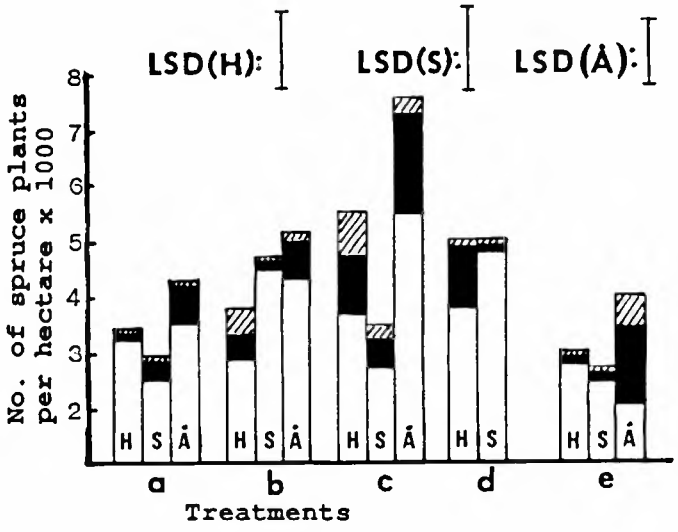

Fig. 2. The effects of various vegetation managements on the number of Norway spruce plants per hectare. Symbols as for Fig. 1

Number. Compared to untreated plots, only treatments b and d gave a significant increase in the number of spruce plants within the experimental period (Fig. 2).

Ås

Height growth: Fig. 1 shows rather uniform plant heights over the whole experimental area at the start of the experiment in 1988. During the subsequent years, only the plants on the overall glyphosate-treated site appeared significantly higher than those on the other sites.

Number: In 1991 only site c showed a significantly higher number of plants (Fig. 2).

Succession of the shrub/herb layer vegetation

Descampsia flexuosa. At Kongliveien, the number of observations made in the year after all the treatments were carried out, appeared somewhat lower than the number on the untreated site (Table 4). After three years, the overall application of glyphosate gave rise to a slightly faster regeneration of the species than the handcutting methods.

The Siljan experiment shows a lower level of *Deschampsia* observations on sites a and b than on the remaining sites, with the lowest level on the clearcut site (Table 5)

At Ås, selective handcutting with stump treatment caused the fastest *Deschampsia* succession (Table 6). On the other experimental sites, the *Deschampsia* coverage appeared relatively similar throughout the experimental period.

Table 4. Succession of shrub/herb vegetation after various vegetation managements (see Table 1). Location: Kongliveien, Hurdal

Plant species	Management	Year of evaluation	No. of obs	% ground cover				
				< 20	21-30	31-40	41-50	>51
				Frequency of observations (%)				
Deschampsia		1989	78	25	16	13	8	38
flexuosa	a	1990	81	30	17	15	9	29
		1991	90	16	23	19	16	26
		1989	76	28	9	9	16	38
	b	1990	55	18	20	26	7	29
		1991	74	24	19	18	11	28
		1989	71	22	18	23	6	31
	c	1990	67	25	13	19	13	30
		1991	82	25	18	14	17	26
		1989	72	18	6	10	20	46
	d	1990	60	21	10	17	10	42
		1991	65	19	15	15	14	37
		1989	88	9	14	17	11	49
	e	1990	86	19	15	14	19	33
		1991	79	11	18	18	16	37
Vaccinium		1989	57	43	21	11	14	11
myrtillus	a	1990	38	54	32	5	3	6
		1991	38	76	21	3	0	0
		1989	66	23	15	12	12	38
	b	1990	66	23	21	21	11	24
		1991	49	39	23	18	10	10
		1989	82	22	6	10	13	49
	c	1990	77	16	19	22	16	27
		1991	69	42	28	16	10	4
		1989	58	10	10	16	28	36
	d	1990	51	30	24	14	8	24
		1991	41	58	15	10	7	10
		1989	79	26	17	18	14	25
	e	1990	80	27	21	16	13	23
		1991	66	35	33	17	8	7
Rubus		1989	44	40	16	7	16	21
idaeus	a	1990	44	29	20	16	14	21
		1991	44	34	25	16	11	14
		1989	22	54	14	9	14	9
	b	1990	21	56	29	10	5	0
		1991	22	50	32	14	4	0
		1989	5	80	0	20	0	0
	c	1990	7	86	14	0	0	0
		1991	8	75	25	0	0	0
		1989	9	45	22	33	0	0
	d	1990	8	62	13	25	0	0
		1991	8	38	25	37	0	0
		1989	9	56	11	11	11	11
	e	1990	14	58	21	14	0	7
		1991	12	67	25	8	0	0

Table 5. Succession of shrub/herb vegetation after various vegetation managements (see Table 1). Location: Siljan, Vestfold

Plant species	Management	Year of evaluation	No. of obs	% ground cover				
				< 20	21-30	31-40	41-50	>51
				Frequency of observations (%)				
Deschampsia		1989	48	25	13	23	19	20
flexuosa	a	1990	63	26	17	11	16	30
		1991	62	10	26	11	14	39
		1989	33	21	25	18	12	24
	b	1990	17	52	24	12	6	6
		1991	23	48	17	18	4	13
		1989	78	10	16	12	14	40
	c	1990	67	24	18	13	21	24
		1991	73	21	20	10	19	30
		1989	72	13	7	13	7	60
	d	1990	74	15	11	21	15	38
		1991	77	16	21	8	14	41
		1989	81	15	7	7	11	60
	e	1990	65	29	15	12	18	26
		1991	72	25	15	14	14	32
Vaccinium		1989	26	53	35	8	0	4
myrtillus	a	1990	30	66	20	7	7	0
		1991	21	86	9	0	5	0
		1989	34	25	24	18	15	18
	b	1990	30	60	13	17	3	7
		1991	18	55	17	11	17	0
		1989	43	44	28	16	2	10
	c	1990	41	64	15	7	2	5
		1991	34	67	9	9	3	3
		1989	50	48	24	20	4	4
	d	1990	49	68	16	14	7	2
		1991	28	82	7	4	3	4
		1989	42	46	19	7	14	14
	e	1990	44	55	25	11	7	2
		1991	34	47	38	6	9	0
Rubus		1989	4	100				
idaeus	a	1990	14	64	29	7	0	0
		1991	44	43	32	9	5	11
		1989	3	100				
	b	1990	7	86	14	0	0	0
		1991	10	70	30	0	0	0
		1989	12	67	25	0	8	0
	c	1990	21	62	33	5	0	0
		1991	26	73	23	4	0	0
		1989	0					
	d	1990	3	100				
		1991	1	100				
		1989	22	44	14	14	14	14
	e	1990	32	53	26	13	3	15
		1991	32	50	22	6	7	15

Table 5 cont.

Plant species	Management	Year of evaluation	No. of obs	% ground cover < 20	21-30	31-40	41-50	>51
				Frequency of observations (%)				
Pteridium		1989	10	50	30	10	0	10
aquilinum	a	1990	20	55	25	5	15	0
		1991	27	30	33	26	7	4
		1989	54	13	9	2	6	70
	b	1990	56	9	5	9	9	68
		1991	57	7	9	7	5	72
		1989	6	33	0	0	17	50
	c	1990	10	60	0	0	0	40
		1991	9	22	34	0	0	44
		1989	10	80	0	10	10	0
	d	1990	8	37	50	0	13	0
		1991	15	33	20	27	20	0
		1989	14	7	30	0	21	42
	e	1990	22	26	14	5	31	24
		1991	20	10	10	20	20	40

Table 6. Succession of shrub/herb vegetation after various vegetation managements (see Table 1). Location: Ås, Akershus

Plant species	Management	Year of evaluation	No. of obs	% ground cover < 20	21-30	31-40	41-50	>51
				Frequency of observations (%)				
Deschampsia		1989	25	32	20	8	12	28
flexuosa	a	1990	29	39	17	3	14	27
		1991	30	37	33	3	17	10
		1989	46	4	4	4	13	75
	b	1990	53	15	20	11	6	48
		1991	47	15	11	13	8	43
		1989	39	15	18	21	22	24
	c	1990	45	22	20	16	18	24
		1991	73	23	13	8	11	45
		1989	20	20	15	20	10	35
	e	1990	25	28	24	20	12	16
		1991	24	25	13	25	17	20
Vaccinium		1989	11	64	0	27	9	0
myrtillus	a	1990	6	100				
		1991	1	100				
		1989	30	44	13	21	13	9
	b	1990	23	66	26	4	4	0
		1991	15	100				
		1989	35	28	17	20	17	18
	c	1990	31	65	16	10	3	6
		1991	24	54	25	9	4	8

Table 6 cont.

Plant species	Management	Year of evaluation	No. of obs	% ground cover				
				< 20	21-30	31-40	41-50	>51
				Frequency of observations (%)				
		1989	7	28	0	29	29	14
	e	1990	14	57	0	29	14	0
		1991	7	43	29	14	14	0
Rubus		1989	10	50	40	10	0	0
idaeus	a	1990	36	49	39	6	3	3
		1991	41	51	29	12	3	5
		1989	10	30	20	20	20	10
	b	1990	9	78	0	11	0	11
		1991	5	60	20	20	0	0
		1989	20	50	20	5	15	5
	c	1990	9	100				
		1991	5	80	20	0	0	0
		1989	18	43	17	11	6	23
	e	1990	14	72	21	7	0	0
		1991	11	73	27	0	0	0
Pteridium		1989	40	32	24	13	15	16
aquilinum	a	1990	36	35	28	14	14	9
		1991	39	18	28	26	10	18
		1989	37	35	8	0	8	49
	b	1990	39	13	15	10	26	36
		1991	44	18	16	20	14	32
		1989	55	29	16	15	15	25
	c	1990	51	27	25	18	12	18
		1991	56	30	25	11	20	14
		1989	50	6	4	8	20	62
	e	1990	55	17	16	15	15	37
		1991	34	15	17	15	12	41
Galeopsis		1989	59	22	10	14	19	35
tetrahit	a	1990	4	100	0	0	0	0
		1991	0					

Vaccinium myrtillus. At Kongliveien all the sites showed approximately the same number of plots with *Vaccinium* the year after the treatments (Table 4). The sites also show a generally declining *Vaccinium* coverage over time. This seems, however, most pronounced after the overall glyphosate and clearcut treatments. The higher number of plots with 10-20% coverage of *Vaccinium* on site a than on site b indicates a stronger influence of the glyphosate application. Similar trends were also found in Siljan (Table 5). But in this experiment clearcutting (b), had an effect on *Vaccinium* similar to that in the glyphosate treatment. The selective handcutting methods have evidently not had an influence on the *Vaccinium* coverage or the succession. Similar reactions were also recorded in the Ås experiment. All the experiments show a decline in occurrence of *Vaccinium*, with the strongest effect on the overall glyphosate site.

Rubus idaeus. Table 4 (Kongliveien) shows a more uniform distribution of *Rubus* on sites

a and b than on the remaining sites, with most of the observation plots showing a 10-20% ground cover of this species. None of the experimental sites show any regeneration of the species.

In Siljan and Ås, the results indicate a comprehensive initial kill as a result of the overall application and a rapid recovery within the experimental period (Tables 5 and 6). The handcut and untreated sites show either no or a declining occurrence of *Rubus* over time.

Pteridium aquilinum. This species was found only in Siljan and Ås. In both areas overall glyphosate application reduced the ground cover of this species, but had only a minor influence on the distribution of the species (Tables 5 and 6).

Galeopsis tetrahit. In the Ås area, this species was found only on the overall glyphosate site (Table 6). The results show an explosively rapid distribution the year after the application, followed by a rapid disappearance.

Broad versus band application of glyphosate

The results from the experiment at Lundbyliveien, Hurdal, are presented in Table 7.

No vegetation. The number of plots free from all vegetation is, as expected, significantly higher after the two glyphosate treatments than on untreated plots. It is, however, highly unexpected to find a lower number of no-vegetation plots and a faster vegetation invasion after overall application than after band application. Three years after the treatments most of the plots with both of the glyphosate treatments show a considerable reinvasion of vegetation.

Bryophyta was slightly stimulated by band application, but the influence was restricted.

Rubus idaeus. Both glyphosate treatments reduced the ground cover of *Rubus*, but the subsequent reinvasion proceeded faster than on untreated areas. The results indicate a slower *Rubus* establishment after the band versus the broad application of glyphosate.

Rubus saxatilis. A decline in occurrence of this species was registered on the untreated site from 1989 to 1991. Both of the glyphosate treatments killed most of the plants, with a subsequent slow recovery after the broad treatment.

Vaccinium myrtillus. On the untreated site, Table 7 shows an increasing coverage from 1989 to 1990 and thereafter a decline. A similar tendency can also be observed on the overall glyphosate plots, though the level of *Vaccinium* coverage appeared higher through all three years than on the untreated plots. Band treatment had evidently a considerably stronger phytotoxic effect on the *Vaccinium* species than did a broad application of glyphosate, and the reinvasion within the experimental period was quite restricted. On all the treatment sites, most of the plots show a mean *Vaccinium* coverage of 10-20%.

Deschampsia flexuosa. On the untreated site the coverage of *Deschampsia* appeared relatively unchanged throughout the experimental period. Broad application of glyphosate caused a considerably faster reinvasion over time than the band treatment. Within the experimental period, the two chemical methods gave a lower ground cover of *Deschampsia* than that on the untreated plot.

Broadleaved graminaea. Band application of glyphosate initially caused a greater reduction of broadleaved grasses than the broad treatment did, but the rate of reinvasion seems somewhat faster after the band treatment. From the second year after the treatments,

no significant differences were found between the treated and untreated sites regarding the coverage of these grass species.

Table 7. The effect of broad versus band application of glyphosate on the succession of the shrub/herb layer vegetation (1: Broad application, 2: Band application, 3: Untreated) Location: Lundbyliveien, Hurdal

Species	Year of evaluation	Management	No. of obs.	% ground cover					
				0	1-20	21-30	31-40	41-50	>51
				Frequency of observations (%)					
No vegetation	1989	1	144	100					
		2	277	100					
		3	11	100					
	1990	1	32	100					
		2	140	100					
		3	20	100					
	1991	1	1	100					
		2	17	100					
		3	1	100					
Bryophyta	1989	1	389	57	30	3	3	3	4
		2	395	65	26	2	2	2	3
		3	295	72	22	2	1	2	1
	1990	1	390	61	31	3	2	2	1
		2	446	71	21	4	2	2	0
		3	295	73	21	2	2	1	1
	1991	1	390	59	32	5	2	2	0
		2	424	71	23	3	1	1	1
		3	234	79	19	1	0	1	0
Rubus idaeus	1989	1	110		67	16	9	5	3
		2	37		70	11	16	3	0
		3	87		43	24	15	7	11
	1990	1	225		50	22	8	8	12
		2	149		60	21	8	6	5
		3	110		36	23	16	8	17
	1991	1	333		21	26	16	14	23
		2	308		34	25	16	7	18
		3	154		33	20	14	13	20
Vaccinium myrtillus	1989	1	52		77	17	2	2	2
		2	0						
		3	29		66	17	7	3	0
	1990	1	64		65	20	8	5	2
		2	4		100				
		3	59		78	14	3	3	2
	1991	1	59		73	17	8	2	0
		2	5		100				
		3	35		66	14	9	8	3
Deschampsia flexuosa	1989	1	44		47	16	9	5	23
		2	53		45	11	17	6	21
		3	195		15	11	11	10	42
	1990	1	87		35	22	20	11	12
		2	29		42	24	7	17	10
		3	152		28	16	13	11	34
	1991	1	149		47	23	16	7	7
		2	84		57	24	14	3	2
		3	174		28	17	13	12	30

Table 7. cont.

Species	Year of evaluation	Management	No. of obs.	% ground cover 0	1-20	21-30	31-40	41-50	>51
				Frequency of observations (%)					
Gram-		1	35		51	17	11	9	12
ineae	1989	2	6		100				
(broad-		3	18		49	39	6	6	0
leaved)		1	94		49	16	14	6	15
	1990	2	97		60	13	11	7	9
		3	101		45	21	16	9	9
		1	194		33	23	17	8	19
	1991	2	232		40	23	15	9	13
		3	171		24	20	13	11	32
Aconitum		1	1		100				
septentrio-	1989	2	1		100				
nale		3	28		53	36	7	4	0
		1	1		100				
	1990	2	0						
		3	30		46	23	17	7	7
		1	1		100				
	1991	2	1		100				
		3	29		76	7	17	0	0
Rubus		1	3		67	33	0	0	0
saxatilis	1989	2	6		100				
		3	15		67	13	7	0	13
		1	7		72	14	0	1	0
	1990	2	20		65	30	5	0	0
		3	3		100				
		1	8		75	25	0	0	0
	1991	2	13		69	23	8	0	0
		3	2		100				
Athyrium		1	0						
filix-femina	1989	2	13		77	15	8	0	0
		3	65		49	22	0	5	2
		1	1		100				
	1990	2	25		72	8	12	8	0
		3	35		57	31	6	3	3
		1	7		100				
	1991	2	41		71	24	3	2	0
		3	45		51	33	16	0	0
* Others		1	25		100				
	1989	2	16		100				
		3	22		100				
		1	54		100				
	1990	2	38		100				
		3	23		100				
		1	33		100				
	1991	2	21		100				
		3	34		100				

* *Rumex acetosella*, *Equisetum* spp., *Oxalis acetosella*. *Veronica* spp., *Epilobium* spp., *Filipendula ulmaria*, *Viola riviniana*

Aconitum septentrionale. A relatively unaltered distribution over time was found on the untreated site, but the ground cover gradually decreased. The two glyphosate treatments show a nearly complete kill, with negligible recovery.

Athyrium filix-femina. Both of the glyphosate treatments displayed a considerable phytotoxic effect and a rather slow recovery after the broad treatment.

Rumex acetosella, *Equisetum* spp., *Oxalis acetosella*, *Epilobium* spp., *Filipendula ulmaria*, and *Viola riviniana* as a group, showed no significant reactions to the two glyphosate treatments.

DISCUSSION

The proportion of hardwoods in spruce regeneration has increased considerably during the last two to three decades (Braathe 1988). Hardwoods, especially *Betula* spp., are serious competitors to Norway spruce at an early stage (Andersson 1984; Braathe 1988; Bärring 1984). *Sorbus aucuparia*, however, is not considered as aggressive as *Betula* because of intense browsing by moose (Hjeljord & Grønvold 1988) and a less competitive morphology (Børset 1985; Lund-Høie & Grønvold 1987). To maintain an ecological balance during the regeneration period, it seems essential to choose methods of vegetation management which reduce the competitive impact of weeds and hardwoods like *Betula*, preserving ecologically important species like *Sorbus* and the diversity of the plant community.

Effects of various vegetation managements

Hardwoods

Betula spp. Overall application of glyphosate appeared generally quite efficient, giving from 90 -100% kill the year after treatment. The rate of regeneration was generally slow and varied between the experimental locations. This was also experienced by Lund-Høie & Grønvold (1987). However, there seemed to be a correlation between the degree of efficiency of the application and the rate of regeneration. At Ås, for example, a 100% initial kill caused only about 1% regeneration three years after treatment. At Kongliveien, the percentages were 2% and 4% and at Siljan 17% and 90%.

That *Betula* regenerates sufficiently after a glyphosate application to give a desired proportion in mixed stands with Norway spruce, is also confirmed by Solbraa & Lund-Høie (1989 and 1990).

The effect of the handcutting methods varied between the experimental locations. At Siljan, for example, the recovery of *Betula* seedlings after clearcutting proceeded more slowly than after glyphosate application. At Kongliveien and Ås, the opposite situation was recorded. However, three years after the treatment at these two locations, the number of seedlings was still less than 20% and mean heights less than 40-50% of those on the untreated sites.

Betula spp. generally shows a high potential of vegetative regeneration. The number of suckers that developed from the stumps in the present experiments appeared to be several times higher than the number of seedlings. Glyphosate treatment on cut stump surfaces reduced the total number of suckers per hectare as well as the mean height of those suckers compared to regenerated seedlings. But the effect varied between the experimental

locations. Stump surface treatment with glyphosate is generally recognized as an efficient method of preventing vegetative regeneration of hardwoods (Lund-Høie & Rognstad 1990), and the method has a highly selective potential.

Selective handcutting resulted in approximately the same number of suckers per stump as that after overall handcutting. This is not in accordance with the general experience that a seedling canopy will reduce the number of suckers, especially at low temperatures (Johansson 1986).

Sorbus aucuparia. Clearcutting had an effect on *Sorbus* similar to that with overall glyphosate application. Excepting the Siljan experiment, the rate of regeneration of seedlings was faster after the glyphosate treatment than after clearcutting. Stump treatment appeared generally more efficient on *Sorbus* than on *Betula*.

Salix spp. showed high sensitivity to glyphosate applied to both foliage and stumps. The vegetative regeneration potential from untreated stumps was rather high. Thus, cutting of *Salix* and also *Sorbus* increased the browsing potential for moose (Hjeljord & Grønvold 1988).

Hardwoods such as *Quercus* spp., *Prunus* spp. and *Sambucus racemosa*, which were also registered in the Ås experiment, showed similar reactions to the various managements. This means that there was a relatively rapid reinvasion after glyphosate application and a high vegetative regeneration potential, which appeared to be suppressed efficiently by glyphosate treatment of the stumps.

All the experiments, independent of species, showed a decline in the number of suckers per stump from 1989 to 1991. The reasons for this are not known, but it may be supposed that competition and intense browsing may have contributed to this process.

Growth reactions of Norway spruce

Lund-Høie & Grønvold (1987) reported a stimulated natural regeneration of Norway spruce after overall glyphosate application. In the present experiments such a reaction was not significant. This was also the case after clearcutting, selective handcutting without stump treatment and on untreated sites. Only selective handcutting combined with stump treatment caused a significant increase in the number of spruce plants from 1989 to 1991 in two of the experiments.

Broad application of glyphosate in 1988 increased the growth throughout the experimental period to a significantly higher level compared with that for the other treatments. This appeared at both the Hurdal and Siljan locations and at Ås after 1990. Similar reactions are also reported by Lund-Høie (1990), and Reynolds et. al. (1989), working with *Picea sitchensis* and *Tsuga heterophylla*.

Release of the spruce plants from competition, especially from *Betula*, is probably the main reason for this growth reaction (Andersson & Bjørkdahl 1984; Bärring 1984, Braathe 1988).

The most relevant alternative to the foliage application of glyphosate in the present experiment, clearcutting, in fact caused a significantly reduced height growth. This was probably due to the tremendous number of suckers on the clearcut sites. According to Andersson & Bjørkdahl (1984), spruce plants need to have a mean height of at least 1.7-2.2 m at the time of cutting to avoid the destructive competition from *Betula* suckers. In the present experiments, none of the plants on the handcut sites had surpassed this minimum

height. A negative growth reaction of Norway spruce after clearcutting of hardwoods is also reported by Lund-Høie (1990).

According to the assumption about the negative influence of *Betula* suckers, treating the stumps with glyphosate in order to suppress the sucker growth should stimulate spruce growth. To some extent, the results in Fig. 1 support this assumption.

Succession of some important forest shrub/herb/grass species

Deschampsia flexuosa is a fairly aggressive weed and rather tolerant to glyphosate at autumn application (Lund-Høie 1975). This was also confirmed in the present experiments (Tables 4, 5, 6). Curiously enough, the clearcutting seems to have reduced the coverage of *Deschampsia* to a greater extent than the glyphosate treatment. The other cutting methods stimulated the species. Generally, *Deschampsia* showed a rather slow revegetation.

Vaccinum myrtillus showed generally a decline in coverage over time, independent of treatment. None of the vegetation managements used had any major influence on this species. The competitive potential of the *Vaccinium* species towards Norway spruce is rather restricted.

Rubus idaeus spreads rapidly in all cases of reduced competition from other shrub/herbs. All the experiments demonstrated a rapid recovery of the species even after a comprehensive initial kill.

Pteridium aquilinum is a rather common weed in forest plantations, with a high vegetative regeneration potential. In the experiment at Ås, only selective handcutting combined with stump treatment of hardwoods increased the coverage of this species.

Galeopsis tetrahit is an annual species which quite often invades a glyphosate-sprayed area, but only for a short period of time. This was also the case in the experiment at Ås. Competitively, this species is of minor importance, but represents an important food source for titmice during the autumn and winter.

Generally, none of the management systems tested had any long-term detrimental effect on the evaluated plant species. Considering both the ecological and silvicultural aspects, foliage application with glyphosate and selective handcutting combined with stump treatment probably represent the best managements methods.

Broad versus band application of glyphosate

As presented in Table 7, the plots that were overall treated with glyphosate were rapidly revegetated. Even after the second growth season there was only a negligible difference between broad-treated and untreated plots. After band treatment, however, the reinvasion of vegetation proceeded much more slowly. The main reason for this is not known, but it may be related to a comprehensive grazing of the bands. Mostly sheep, but also cattle, followed the bands while grazing. It is also possible that the bands may have functioned as brooklets during periods of heavy precipitation.

Compared to untreated plots, *Bryophyta* has evidently been stimulated by the glyphosate applications, with just minor differences between the two methods of application. This was also the case regarding *Rubus idaeus*, and this species occurred more frequently on the overall treated area than on the band-treated area.

Vaccinium myrtillus was evidently stimulated by the broad application, but the coverage was greatly reduced and the recovery highly retarded after the band treatment.

A similar trend was found for *Deschampsia flexuosa*. The coverage of broadleaved grasses was also highly reduced by the glyphosate treatments, mostly by the band treatment. A rapid revegetation was observed after both treatments, however.

Aconitum septentrionale displayed a high sensitivity to glyphosate and a slow recovery. This was also true for *Rubus saxatilis* and the fern *Athyrium filix-femina*. However, both species recovered faster on the bands than on the overall treated area.

The spread of species like *Rumex acetosella*, *Equisetum* spp., *Filipendula ulmaria* and *Viola riviniana* was stimulated after both modes of glyphosate application.

Regarding sensitivity of the main species involved, the present results are supported by earlier findings that no species is eradicated by any of the vegetation managements used (Lund-Høie & Grønvold 1987). Where there was reduced occurrence shortly after the treatments, the revegetation appeared more or less rapid. Neary et al. (1990), using several herbicides annually over several years, found a strong influence on plant diversity. However, two applications, with sulphometuron followed by glyphosate or triclopyr caused only a small reduction in diversity. Consequently, there is no reason to believe that just one glyphosate treatment per 60-100 years has any detrimental effect on the long-term regeneration of forest plant species, nor on their diversity. The same conclusion may be drawn for the handcutting managements (Kimmins et al., 1989). Swindel et al. (1987) came to the same conclusion regarding the effects of intensive site preparation treatment.

SUMMARY

In this paper the effects of various vegetation management methods and their impact on major forest species including Norway spruce are analysed. The management methods involved were overall glyphosate application, manual clearcutting of all hardwoods and selective handcutting of *Betula* spp. leaving a considerable amount of *Sorbus aucuparia* with or without glyphosate stump treatment. An untreated site was used as reference area. The experiments were run at three different locations, Hurdal and Ås in the county of Akershus and at Siljan, in Vestfold. In a fourth experiment, broad and band applications of glyphosate were compared regarding succession of plant species after the treatments. The experiments ran from 1988 to 1991, and were evaluated in August-September each year starting in 1989.

The parameters investigated were succession of hardwood seedlings, and development of stump suckers after the various vegetation managements. The experiments also evaluated the impact on and succession of shrub/herb species and the effect of the managements on establishment and growth of Norway spruce.

The evaluations were based on 2 x 2 m or 0.5 x 2 m plots at given spacing.

The results showed that no particular one of the management methods involved had a detrimental impact on the long-term diversity of the plant community characterizing the experimental areas. The most effective treatments in this respect appeared to be overall glyphosate application and manual clearcutting. However, the recovery of controlled species was relatively speedy. The impact on vegetation appeared more pronounced after band than after broad application of glyphosate. Selective cutting of especially *Betula* spp. mixed with *Sorbus aucuparia*, and combined with chemical treatment of the stumps, optimized the

growth conditions for the ecologically important *Sorbus* species. In contrast to clearcutting of the hardwoods, the selective management referred to also favoured the growth of Norway spruce. However, the most pronounced growth reactions on Norway spruce were found on the overall glyphosate-treated plots.

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