POLLEN FORMED UNDER POLLUTION AFFECTS SOME QUANTITATIVE CHARACTERS OF SCOTS PINE (*PINUS SYLVESTRIS* L.) SEEDS

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ABSTRACT

The effect of polluted Scots pine pollen on some seed quantitative characters was studied by control crossings on one mother clone in Kórnik done with pollen of three geographically distant populations growing in two sites: (1) polluted, in the vicinity of mineral fertiliser factory (Luboń) and (2) control, relatively free of pollution (Kórnik). The pollination with pollen from Luboń caused a significant loss of 1000 filled seeds weight (p = 0.024) and mean cone weight (p = 0.103). The results obtained implicate some after-effects resulting from the pollen formation environment. Implications are outlined shortly in the paper..

Key words: *Pinus sylvestris*, environmental pollution, controlled crosses, seed quality, after-effects.

INTRODUCTION

The environmental impacts on generative reproduction processes in coniferous trees are well documented in Scots pine. It is commonly accepted that adaptation of tree progenies depends also on the site conditions under which the process of sexual reproduction was performed in (*e. g.* JOHNSEN 1989 a and 1989 b, JOHNSEN & ØSTRENG 1994, SKRØPPA *et al.* 1994). Parental environment also affected development of Scots pine progeny *e. g.* seed weight and height of seedlings (DORMLING & JOHNSEN 1992). There is evidence that these factors may cause some genetic selection in the progeny too (DORMLING & JOHNSEN 1992).

Between-population gene flow through long distance pollen transport may cause similar effects. Migrating pollen may effectively contribute in pollination and fertilisation (KOSKI 1970) and may implicate some genetic consequences in the adaptation process of forest trees (LINDGREN *et al.* 1995). Contamination of pollen in seed orchards is another phenomenon in which long-distance transport of pollen is involved (NAGASAKA & SZMIDT 1985, YAZDANI & LINDGREN 1991, BURCZYK 1992). This outside pollen may influence genetic variation of seed orchard progeny (MÜLLER-STARCK 1991). A similar situation could very likely be in the case of pollen formed at a polluted site and then transported for long distances to relatively unpolluted stands or seed orchards.

Most experiments aimed at study of the influence of external pollutants on pollen vigour and fertilisation capacity (COX 1992). According to our knowledge no information is available on the influence of pollutants deposited inside pollen grains formed in a polluted environment on pollen vigour. Recently OLEKSYN *et al.* (in press) noticed significantly higher level of S, Mn, Al, Na, Cu, Ni and Cd in Scots pine pollen of different populations at a polluted site compared to a control one. An effect of the higher level of „endogenous" pollutants on pollen vigour and fertilisation ability seems thus to be very plausible.

One can thus expect that such pollen transported for long distances may also influence quality of progeny in the affected nonpolluted populations. Such an assumption inspired the experiment with control crossings which aimed to study the effect of the environment under which Scots pine pollen was formed on the quality of seeds obtained. This aim was expected to be achieved by finding differences between seeds obtained from control crosses using different male parents from a polluted and a control site mated with a genetically homogenous female parent.

MATERIAL AND METHODS

In order to examine this question, controlled crosses were performed with the pollen collected at two sites: (1) under pollution in the vicinity of a mineral fertiliser factory (Luboń) and (2) control, relatively free of pollution (Kórnik). In both sites the provenance trial with the same set of 20 European Scots pine populations was established in 1984 (OLEKSYN & BIAŁOBOK 1986,

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Table 1. Geographic origin of male parental populations

Country	Provenance	Population no.	Latitude N	Longitude E
Russia	Serebryanskoe	3	58° 50'	29° 07'
Poland	Rychtal	8	51° 08'	17° 55'
Slovakia	Záhorie	16	48° 46'	17° 03'

Table 2. Germination ability of pollen in 1996 test

Site	Populations		
	Serebryanskoe	Rychtal	Záhorie
	%		
Control (Kórnik)	66.7	23.8	77.8
Polluted (Luboń)	56.0	22.4	60.6

OLEKSYN 1988). Male strobili were collected in May 1994 at both sites from the same three populations (Table 1). The male flowering in 1994 was rather poor and a collection of male strobili was done on all flowering trees from each population in Kórnik and in Luboń (figures in brackets), *i.e.* 3(4) trees for Serebryanskoe (Russia), 5(4) trees for Rychtal (Poland) and 7(3) trees for Záhorie (Slovakia). Thus the pollen used for cross pollination was a multi tree mixture from each male population.

Pollen was extracted indoors under electric light at a temperature of around 25–28 °C. After one day drying in a $CaCO_3$ dessicator, pollen was used for controlled crossings which were done by using Polish clone of Scots pine No. K–11–03 as female parent. This clone was selected because it was prolific bearing in 1994 even 8 female strobili per shoot. It originates from the Kampinos Natural Forest (central-eastern Poland) and grows in a second generation seed orchard in Kórnik. Ten grafts of the mentioned clone were selected for the experiment thus the each crossing was replicated tenfold. Six branches on each maternal graft were selected and female strobili on each branch were pollinated with a pollen mixture of one out of six selected male populations. No viability test was done because of limited amounts of pollen from some populations. Such a test was realized in 1996 (Table 2). Mature cones were collected in February 1996 and seeds were extracted carefully in the laboratory. An analysis of variance (JMP version 3.0.2., SAS Inst. Inc.) was used for statistical evaluation of several cone and seed yield parameters (Table3). One maternal graft growing poor in 1995 died shortly after cone collection and thus only nine grafts were evaluated. Percentage values were subjected to the arcsine transformation.

RESULTS AND DISCUSSION

Table 3 contains results of variance analysis of different female cone and seed characters. Less control-pollinated female strobili survived after pollination with pollen from polluted site (Luboń) than from control one (Kórnik) (64.2% and 70.4% respectively), but the difference was not statistically significant ($p = 0.1982$). Weight of 1000 filled seed was significantly influenced by the site of pollen formation ($p = 0.024$). A thousand filled seeds produced after control pollination with pollen originating from the polluted site (Luboń) were 9.09% lighter than seeds resulting from pollination with pollen from the control site in Kórnik (8.913 g and 9.804 g respectively). It is hard to compare this result with others, *e. g.* that obtained by PALOWSKI (1994) who found open-pollinated seeds formed under pollution in the Upper Silesia much heavier than those from control unpolluted plot located several hundreds of kilometres north-east, in the Bialowieża Primeval Forest. However such effect may reflect also the differences between populations of unknown origin and population × site interaction.

The loss of 1000 filled seed weight found in this experiment is an important quantitative effect of pollen formed under pollution and this means that progeny seeds of males influenced by pollution have less content of metabolites which may diminish the growth potential at the beginning of seedlings growth. GIERTYCH (1974) found a significant positive correlation between seed weight and height growth of Scots pine seedlings even up to age 16. The difference in seed weight was also found to be responsible for differences in height growth

Table 3. Results of variance analysis of various quantitative cone and seed characters

Parameter analysed	Source of variation										
	Total	Localisation (L)			Provenance of males (M)			M × L			Residual
	df	df	F	P > F	df	F	P > F	df	F	P > F	df
Female strobili survival, %	49	1	0.803	0.375	2	0.721	0.420	2	0.169	0.845	44
Mean dry weight of one female cone, g	46	1	2.781	0.103	2	1.009	0.373	2	0.468	0.630	41
Mean number of seeds (filled + empty) per cone	46	1	1.781	0.189	2	1.737	0.189	2	0.942	0.398	41
Mean number of filled seeds per cone	46	1	0.071	0.791	2	1.413	0.255	2	0.370	0.693	41
Mean weight of 1000 (filled + empty) seeds per cone, g	46	1	1.380	0.247	2	0.170	0.844	2	0.009	0.991	41
Mean weight of 1000 filled seeds, g	45	1	5.483	0.024	2	0.889	0.420	2	0.310	0.735	40
% of filled seeds	46	1	0.056	0.811	2	0.009	0.990	2	0.207	0.814	41

df – degrees of freedom; F – F-test; P > F – level of statistical signifcance

of Scots pine seedlings after identical crosses were done at different latitudes (DORMLING & JOHNSEN 1992).

Less significant difference ($p = 0.103$) was noticed between mean weight of a cone from the female clone pollinated with pollen from Luboń (10.57 g) vs. pollen from Kórnik (11.90 g).. Presence of pollen grain on ovules is necessary for female strobili growth and the negative effect of polluted pollen on cone weight is thus likely. MAMAEV & SHKARLET (1972) also noticed loss of weight of Scots pine cones under pollution but this effect was a result of the influence of pollutants on both female and male generative organs. My observation refers only to the influence of polluted pollen affecting female strobili development at a nonpolluted site.

Pollen of the studied Scots pine populations was also analysed chemically and biochemically. As was shown by OLEKSYN *et al.* (in press), the accumulation of such elements as S, Al, Mn, Cu, Ni and Cd was significantly higher in pollen at the polluted site in Luboń than in the control one in Kórnik, and this may potentially adversely affect its function. It was also observed that the level of free proline was 32% lower in the pollen formed in the polluted area (Luboń) compared to the control one (Kórnik) and this could be a result of some disturbances in the translocation of proline during pollen grain maturation (ŻYTKOWIAK *et al.* 1996). On the other hand the content of total phenols and orthodiphenols was higher by 111% and 27% respectively in pollen formed under pollution than in that coming from the control plot (ŻYTKOWIAK *et al.* 1996). PUKACKI & CHAŁUPKA (1995) found lower levels of total phospholipids and unsaturated fatty acids in Scots pine pollen from the polluted area, which indicates a process of degradation in the cell wall of pollen grains. There is no direct data on the relationship between observed biochemical and biophysical changes in pollen under pollution and their influence on fertilisation process and seed quality, but such a hypothesis cannot be rejected.

The preliminary results discussed above implicate Scots pine pollen formed in a polluted environment with some negative impact on seed quality. It can be said that long distance transport of such pollen may result in diminishing value of seed yield produced in a relatively non-polluted stands and seed orchards. It is likely that this will reflect also in some qualitative seed characters and further analysis of subject will be carried on with the same seed material.

These preliminary results should be verify by addi-

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tional controlled crosses using a set of male parents mated with more than one female parents.

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