

A study on the variation of morphological characteristics of silver fir (*Abies alba* Mill.) seeds and their internal structure determined by X-ray radiography in the Beskid Sądecki and Beskid Niski mountain ranges of the Carpathians (southern Poland)

K. SKRZYSZEWSKA, J. CHŁANDA

Department of Forest Trees Breeding, Faculty of Forestry, Agricultural University of Cracow, Cracow, Poland

ABSTRACT: The variation of the morphological characteristics and internal structure of silver fir (*Abies alba* Mill.) seeds from 4 stands growing at the altitude of 625–750 m in the Beskid Sądecki and Beskid Niski mountain ranges of the Carpathians in southern Poland was estimated. Seeds were collected in the second half of September 2004. The weight of 1,000 air-dry seeds was determined and their viability was estimated with the use of X-ray radiography. The length and width of the embryo, endosperm and embryo cavity were measured on X-ray photographs. The morphological seed characteristics, i.e. length, width, volume, surface area, and wing colour (light-brown, brown, dark-brown, and cherry-red) were also included in the analysis. The statistically significant effect of the provenance on the parameters of the embryo, endosperm and embryo cavity as well as on the occurrence of full and insect-attacked seeds was found. The weight of 1,000 seeds was below 55 g which is considered to be a long-term mean for silver fir in Poland. It was positively correlated with the length and width of the embryo, endosperm and embryo cavity as well as with the degree of filling of the embryo cavity with the embryo. The weight of seeds was also positively correlated with their length, width and surface area. It was found that the wing colour was correlated with the weight of 1,000 seeds as well as with the proportion of insect-attacked and empty seeds. The proportion of seeds with brown wings was negatively correlated with the weight of 1,000 seeds and numbers of full seeds, while it was positively correlated with numbers of seeds infested by insects and empty seeds. The occurrence of light-brown and cherry-red wings was associated with a high proportion of full seeds. The results of the correlation between seed parameters and selected characteristics of test trees indicated statistically significant relationships. A positive correlation was found between the proportion of full seeds and the height, crown shape, dbh, and bark thickness of trees while the correlation between these tree characteristics and the proportion of insect-attacked seeds was negative.

Keywords: *Abies alba*; X-ray; seed viability; internal seed structure; seed morphological characteristics; Carpathians

Rational forest management requires an adequately large and properly managed seed base which would provide a sufficient amount of high-quality seed. For silver fir (*Abies alba* Mill.), stands of the Beskid Sądecki and Beskid Niski mountain ranges of the Carpathians, belonging to the most valuable provenances of this species in Poland,

make such a seed base. They are of a high genetic and silvicultural value, and their progeny is characterized by good growth and high survival as well as high plasticity, i.e. weak response to variable climatic and soil conditions, and good resistance to environmental pollution (SKRZYSZEWSKA 1999, 2007).

Under natural conditions silver fir produces an abundant amount of seeds every 3–4 years. However, in mountains a good seed year occurs every 5 to 8 years (ZAŁĘSKI, KANTOROWICZ 1993). To ensure the continuity of planting stock production, also during poor seed years, it is necessary to store seed for long periods of time. Such seed should be characterized by the high germination potential determined mainly by the capacity and energy of germination.

The relatively low germination capacity of silver fir seeds as well as small variation of physical characteristics of viable and unviable seeds, making their grading difficult, cause that a complex appraisal of their quality would have to be complemented with estimations, also using the analysis of internal seed development, on the basis of which it will be possible to improve their grading. Practically, there are no detailed elaborations concerning the biological variation of silver fir seeds. Some studies, as for example that of LAFFERS (1979), showed inter-provenance diversification of seed weight of silver fir of different provenances. Also in the experiment of FOBER (1984) the weight and size of seeds differentiated the investigated silver fir provenances. A considerable diversification of these characteristics was shown by the Carpathian provenances as well as the provenances from the Bohemian-Moravian Highlands.

The results obtained by SABOR (1984) showed that the probability of seed germination increases with an increase in their weight. According to this author seeds of silver fir up to 40 mg in weight (at 15% moisture) are characterized by low sowing quality, not exceeding 20%. When analyzing the internal structure of black pine seeds, ZAŁĘSKI and BORKOWSKA (1993) found that seeds with the underdeveloped embryo germinated at a lesser per cent and produced seedlings of worse quality. Studies concerning seeds of Weymouth pine confirmed that the sowing success and growth of 1-year-old seedlings depend on the class of embryo development and its length (ZAŁĘSKI 1990). The development of the embryo and endosperm in Scots pine and Norway spruce was presented by GOZDALIK (1999). This author observed a significant positive relationship between the growth of 1-year-old seedlings and the embryo length as well as length and width of the endosperm. The effect of the embryo and endosperm size of tree seeds as well as that of seed quality determined by X-ray radiography were also studied by SIMAK and GUSTAFSSON (1954), KAMRA (1972, 1976), BELCHER (1974), SIMAK (1974, 1980), and SMIRNOVA (1978). In his study on significantly diversified provenances of *Pinus cabaea* var. *hondurensis*, SALAZAR (1986) found a significant effect of seed weight and length

Table 1. Characteristics of silver fir parent stands in which seeds were collected

Location	Characteristics of parent stands			
Forest range	Powroźnik	Kopciowa	Berest	Feleczyn
Sub-compartment	115 b	10 a	151 b	348 a
Forest administrative unit	Krynica Forest Experimental Station		Nawojowa Forest District	
Area (ha)	17.85	15.00	3.51	13.46
Natural forest province	Province 6; Gorce and Beskid Sądecki		Province 7; Beskid Niski	
Latitude	20°96'E	20°96'E	20°57'E	20°50'E
Longitude	49°37'N	49°48'N	49°33'N	49°28'N
Altitude (m)	625–750	690–720	575–675	525–625
Age (years)	130	95	110	100
Species composition	<i>Abies alba</i>	<i>Abies alba</i> – <i>Fagus sylvatica</i>	<i>Abies alba</i>	<i>Abies alba</i>
Forest site type	mountain forest			
Configuration	medium height mountains	medium height mountains	medium height mountains	medium height mountains
	gentle slope, S-E exposure	gentle slope, S exposure	steep slope, S exposure	steep slope, S-E exposure
Ground vegetation	grass, blackberry, lady fern, male fern	grass, blackberry, lady fern, male fern	grass, lady fern, male fern	grass

on seed germination and growth of seedlings under controlled conditions in plastic tunnels and no effect of other seed characteristics determined by X-ray radiography. The effect of seed aging on their quality was analyzed by MACHANIČEK (1981).

In the study presented in this paper, the variation of size and internal structure of silver fir seeds, collected in the most valuable stands of this species in the Beskid Sądecki and Beskid Niski mountain ranges, was analyzed. The knowledge of seed size and of development of the embryo and endosperm of silver fir seeds will permit to estimate their viability prior to collection in seed stands.

MATERIAL AND METHODS

The silver fir seeds tested during this study originated from four stands of confirmed genetic and silvicultural quality, selected in the Beskid Sądecki and Beskid Niski mountain ranges. Their location and characteristics are presented in Table 1.

The cones were collected between 16 and 29 September 2004 from 50 test trees in each stand. In addition, seeds were collected from 14 plus trees in the Powroźnik Forest Range (sub-compartment 155b, Beskid Sądecki). The cones were stored at the constant temperature of 25°C for the period of 2 months. After seed extraction the estimation of variation of the morphological characteristics and internal structure of silver fir seeds was started on 2–4 December 2004. In the case of two stands (Kopciowa, sub-compartment 10a and Powroźnik, sub-compartment 115b) the analyses were carried out for provenances and families because seeds were collected from single marked trees. The analysis of the investigated characteristics was carried out according to the rules obligatory in Polish State Forests (ZAŁĘSKI et al. 2000).

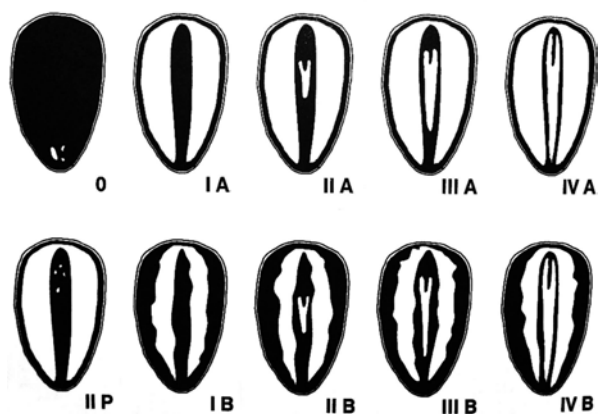


Fig. 1. Diagram of development of the embryo and endosperm according to Swedish classification (ZAŁĘSKI et al. 2000)

Weight of 1,000 air-dry seeds was determined on the basis of the arithmetic mean of 3 replications, 100 seeds each, representing one test tree, multiplied by 10. These seeds were taken from the fraction of pure seeds and weighed exact to 0.001 g.

The **viability of silver fir seeds** was determined using X-ray radiography. From each test tree 200 seeds (4 replications of 50 seeds each) were X-rayed. X-ray photographs were taken at the Forest Research Institute in Warsaw using the ISB-40 apparatus. Seeds were placed on perforated templates. Each X-ray photograph was numbered permitting its identification. Two groups of seeds were distinguished on the basis of X-ray photographs: a) viable – fully developed and able to germinate, and b) unviable – empty, with underdeveloped embryo and endosperm, or infested by insects.

The **development of the embryo and endosperm** was estimated on the basis of the X-ray photographic image. Seed photographs were examined under magnification of 20×, using the Minox K6 apparatus. Seed embryos were classified according to the Swedish scale, and then percentages of seeds in individual developmental classes were determined (ZAŁĘSKI et al. 2000).

The classification used in this study included the following classes of the embryo and endosperm development (Fig. 1):

- A – the endosperm is fully developed, and almost fully fills the seed space;
- B – the endosperm is not fully developed, and does not fully fill the seed space, it is frequently undulated and deformed;
- 0 – the seed is empty;
- I – the seed has the endosperm and embryo cavity but no embryo;
- IIP – the seed has the endosperm and one or more tiny punctual embryos;
- II – the seed has the endosperm and one or more embryos but neither of them is larger than a half of the embryo cavity;
- III – the seed has the endosperm and one or more embryos but neither of them is smaller than a half and larger than three fourths of the embryo cavity;
- IV – the seed has the endosperm and one embryo developed fully, which fully or almost fully fills the embryo cavity.

The obtained results permitted to determine the **potential seed germination capacity** using the universal formula for coniferous species (ZAŁĘSKI et al. 2000):

$$Z = \frac{0.5 \times N_2 + N_3 + N_4}{N} \times 100\%$$

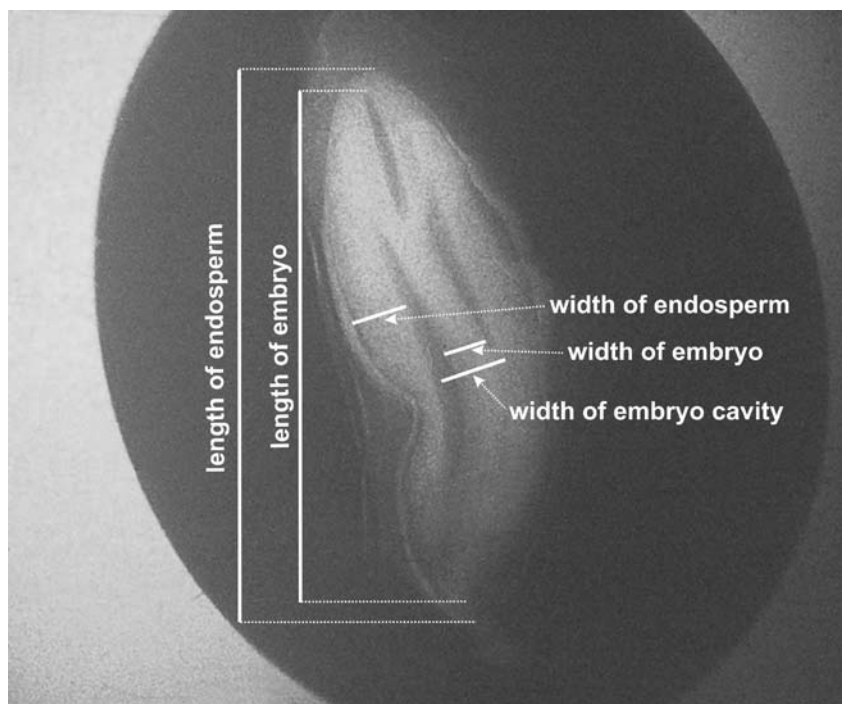


Fig. 2. The X-ray photograph of the silver fir seed with marked places of measurements of its internal structure characteristics

where:

N_2, N_3, N_4 – seed numbers in embryo development classes II, III, and IV,

N – total number of seeds in a sample.

Sizes of the embryo, endosperm and embryo cavity were measured with a millimetre scale under magnification of 20 \times , to the nearest 0.05 mm, using the Minox K6 projector.

The following measurements were made on X-ray photographs: embryo length, embryo width at $\frac{1}{2}$ of its length, endosperm length, endosperm width at $\frac{1}{2}$ of its length, length of the embryo cavity and its width at $\frac{1}{2}$ of its length (Fig. 2).

The **morphological** characteristics of silver fir seeds, i.e. length, width, volume, and surface area of a seed as well as colour of its wing, determined during earlier studies (BARAN 2005), were also included in the correlation analyses.

The results were analyzed statistically using MS Excel 2003 and the Statistica 6.0 PL program. The variance analysis of the investigated characteristics at the level of provenances and families was checked

by the Brown-Forsyth test. The significance of differences was determined at the level $p \leq 0.001$. In all analyzed correlations the level of statistical significance $\alpha = 0.05$ was assumed.

RESULTS

Weight of 1,000 seeds

The weight of silver fir seeds collected in stands under investigations was relatively small. In all provenances the weight of 1,000 seeds did not reach 55 g, considered to be a long-term mean for silver fir in Poland (BODYL 2005).

The highest weight was reached by seeds from permanent seed stands in Feleczyn and Berest Forest Sections, i.e. 53.27 and 51.20 g, respectively. The mean weight of 1,000 seeds collected in the permanent seed stand in Powroźnik Forest Range was 49.41 g (Table 2). Plus trees in this stand produced seeds lighter than the seed mean weight for the stand. Their mean weight was 46.47 g.

Table 2. Mean weight of 1,000 seeds of silver fir of various provenances

Characteristic	Value	Powroźnik		Kopciowa	Berest	Feleczyn
		stand	plus trees			
Weight of 1,000 seeds (g)	mean	49.41	46.47	38.92	51.20	53.27
	stand. dev.	8.18	7.40	9.24	–	–
	CV (%)	16.55	15.92	23.94	–	–

Table 3. Viability of silver fir seeds determined on the basis of the X-ray photographic image of the internal seed structure

Seeds		Powroźnik		Kopciowa	Berest	Feleczyn
		stand	plus trees			
Viable	able to germinate (%)	43.9	43.1	15.8	44.0	42.0
	empty (%)	23.8	21.1	27.3	18.0	26.0
Unviable	with underdeveloped endosperm/embryo (%)	27.0	30.9	31.2	30.0	32.0
	infested by insects (%)	5.3	4.9	25.7	8.0	0.0

Table 4. The analysis of variance of silver seed viability determined on the basis of the X-ray photographic image showing changes in the internal seed structure

Characteristic		<i>F</i> -test	Significance level
Viable seeds	able to germinate	15.578	< 0.001
Unviable seeds	empty	2.010	0.159
	with underdeveloped endosperm/embryo, infested by insects	9.246	0.003

However, they were more homogeneous than seeds from the entire sub-compartment 115b, as indicated by the values of coefficients of variation, i.e. CV = 15.92% for plus trees and 16.55% for the whole stand.

A high disproportion between the weight of seeds originating from the Kopciowa Forest Range and the remaining provenances is worthy of notice. The mean weight of 1,000 seeds from Kopciowa was only 38.92 g. This population was also most diversified as expressed by the coefficient of variation equal to 23.49%.

Viability of silver fir seeds

According to the regulation of the Polish Minister of Environmental Protection of 18 February 2004, concerning the forest breeding material, the viability

of silver fir seed should not be lower than 20%. This threshold value was exceeded by seed from all stands with the exception of Kopciowa (sub-compartment 10a) where it was 15.8%. Seeds from the remaining stands were characterized by even and relatively good viability, i.e. 44.0% for the Berest Forest Range, 43.9% for the Powroźnik permanent seed stand, 43.1% for Powroźnik plus trees, and 42.0% for the Feleczyn Forest Range (Table 3).

The one-way analysis of variance showed a significant effect of the provenance on the occurrence of full seeds as well as on the occurrence of seeds infested by insects. At the same time this analysis excluded the effect of the provenance on the number of empty seeds (Table 4).

Percentages of seeds in the embryo development classes (acc. to Swedish classification) and the potential germination capacity of seeds of analyzed silver fir provenances are shown in Table 5.

Table 5. Percentage of seeds in individual embryo development classes and the potential germination capacity of silver fir seeds

Provenance		Embryo development classes (%)					Seeds infested by insects (%)	Potential germination capacity (%)
		I	II	III	IV	empty		
Powroźnik	stand	27.0	0.0	0.0	44.0	24.0	5.0	44.0
	plus trees	31.0	0.0	0.0	43.0	21.0	5.0	43.0
Kopciowa		31.0	0.0	0.0	16.0	27.0	26.0	16.0
Berest		30.0	0.0	2.0	42.0	18.0	8.0	44.0
Feleczyn		32.0	0.0	0.0	42.0	26.0	0.0	42.0

Table 6. Mean values of the length and width of the embryo, endosperm, and embryo cavity, and the embryo cavity filling per cent in seeds of silver fir of various provenances

Characteristic		Powroźnik		Kopciowa	Berest	Feleczyń
		stand	plus trees			
Length (mm)	embryo	10.58	10.29	9.85	10.31	10.71
	endosperm	11.43	11.13	10.92	11.28	11.71
	embryo cavity	11.12	10.84	10.61	11.06	11.41
Width (mm)	embryo	0.92	0.92	0.91	0.86	1.05
	endosperm	1.08	1.13	1.06	1.02	1.03
	embryo cavity	1.51	1.50	1.57	1.47	1.57
Filling of the embryo cavity (%)		58.47	58.47	54.20	55.29	63.13

Length of the embryo, endosperm, and embryo cavity

Values of the mean length of the embryo, endosperm, and embryo cavity of silver fir seeds are presented in Table 6.

Seeds from the Feleczyń Forest Section were characterized by the longest embryo, endosperm, and embryo cavity. Next positions in this respect were assumed by seeds from the Powroźnik stand, Berest stand, and Powroźnik plus trees. Seeds from the Kopciowa stand had the distinctly shorter embryo, endosperm, and embryo cavity.

Width of the embryo, endosperm, and embryo cavity

Width of the embryo, endosperm, and embryo cavity did not show such a distinct arrangement of mean values as it was the case with their length.

The greatest mean *width of the embryo* was found for seeds originating from Feleczyń, i.e. 1.05 mm. Mean values of this characteristic for seeds from Powroźnik was 0.92 mm, from the entire stand as well as from plus trees, and 0.91 mm for seeds from Kopciowa. The smallest width was found for seeds from Berest, i.e. only 0.86 mm (Table 6).

Seeds from Powroźnik turned out to be least variable in respect of the embryo width. The calculated coefficient of variation in their case was 8.84%, while seeds from Kopciowa (sub-compartment 10a) were almost twice as variable (CV% = 14.63).

The greatest mean *width of the endosperm* was found for seeds from the permanent seed stand in Powroźnik, i.e. 1.08 mm. This value was 1.13 mm for plus trees from Powroźnik, 1.06 mm for Kopciowa, 1.03 mm for Feleczyń, and 1.02 mm for Berest (Table 6). In respect of this characteristic seeds from

Kopciowa were most diversified (CV% = 15.74), while those from Powroźnik were least diversified (CV% = 9.56).

The *width of the embryo cavity* turned out to be the most balanced characteristic of the internal structure of silver fir seeds. Its mean value ranged from 1.47 mm for seeds from Berest to 1.57 mm for seeds from Kopciowa and Feleczyń. The coefficient of variation ranged from 6.57% (Powroźnik) to 9.32% (Kopciowa).

The embryo cavity filling per cent pointed to the existence of the relationship between the size of the embryo and the degree of filling of the embryo cavity space.

The highest values of this characteristic were found for seeds from Feleczyń (63.13%) and Powroźnik (58.47%), while the lowest ones for seeds from Berest (55.29%) and Kopciowa (54.20%). The coefficient of variation varied from 6.44% for Powroźnik plus trees to 12.32% for the Kopciowa stand (Table 6).

The analysis of variance showed the statistically significant effect of the stand (provenance) on the length of the embryo, endosperm, and embryo

Table 7. Analysis of variance of the length and width of the embryo, endosperm, and embryo cavity of silver fir seeds from tested stands

Characteristic		F-test	Significance level
Length	embryo	18.518	< 0.001
	endosperm	8.785	< 0.001
	embryo cavity	9.172	< 0.001
Width	embryo	3.952	0.008
	endosperm	0.740	0.528
	embryo cavity	5.387	0.001

cavity as well as on the width of the embryo and embryo cavity (Table 7). No significant differences were found between provenances in respect of the endosperm width.

CORRELATIONS

The analysis of correlations between the characteristics of the internal seed structure and seed morphological and qualitative characteristics showed their statistical significance. A strong positive correlation was found between the weight of 1,000 seeds and internal seed structure characteristics, i.e. length and width of the embryo, endosperm, and embryo cavity. The correlation between the embryo cavity filling and the seed weight was statistically insignificant. This study has confirmed the dependence of 1,000 seed weight on the degree of seed filling. There was a positive correlation in the case of full seeds and a negative one in the case of empty seeds and seeds infested by insects (Table 8). The weight of 1,000 seeds was also positively correlated with seed length and width as well as with its surface area and all analyzed characteristics of seed internal structure (Table 9).

The relationship between the occurrence of seed wings of different colours (light-brown, brown, dark-brown, and cherry-red) and the weight of 1,000 seeds as well as percentages of seeds infested by insects and empty ones was also determined. The proportion of dark-brown wings was distinctly negatively correlated with the weight of 1,000 seeds as well as with the numbers of full seeds, while it was positively correlated with the numbers of seeds infested by insects and empty ones ($r = -0.374, -0.578, 0.428$, and 0.315 , respectively). The occurrence of wings of light-brown and cherry-red colours indicated the predominance of full seeds.

When analyzing the correlation between seed parameters and selected characteristics of parent trees, a statistically positive correlation was found between the proportion of full seeds and the height, crown shape, dbh, and bark thickness of trees. On the other hand, the correlation between these tree characteristics and the proportion of empty as well as insect-infested seeds was negative (Table 10).

DISCUSSION

This study concerning the biometric estimation and development of the internal structure of silver fir seeds collected in 2004 in four Carpathian stands of high genetic and silvicultural value showed that they were characterized by a relatively small variation of 1,000 seed weight and a high diversification

Table 8. Coefficients of correlation of the characteristics of silver fir seeds. Significant values (at $\alpha = 0.05$ level) are bold

Characteristics of seed internal structure	Length			Width		Filling of		Seeds	
	embryo	endosperm	embryo cavity	embryo	endosperm	embryo cavity	embryo cavity	full	infested
Length	embryo	-							
	endosperm	0.967	-						
	embryo cavity	0.972	0.996	-					
Width	embryo	0.532	0.468	0.486	-				
	endosperm	0.539	0.526	0.541	0.734	-			
	embryo cavity	0.481	0.468	0.472	0.809	0.627	-		
Filling of embryonic canal		0.435	0.290	0.310	0.649	0.409	0.128	-	
	able to germinate	0.480	0.383	0.386	0.121	0.205	0.031	-	0.325
Seeds	insect infested	-0.327	-0.232	-0.251	-0.017	-0.133	0.020	-0.745	-
	empty	-0.424	-0.385	-0.372	-0.172	-0.207	-0.072	-0.727	0.113

Table 9. Coefficients of correlation between the morphological characteristics and the characteristics of the internal structure of silver fir seeds. Significant values (at $\alpha = 0.05$ level) are bold

Morphological characteristics of seeds (according to BARAN 2005)	Length			Width			Filling of embryo cavity		Seeds	
	embryo	endosperm	embryo cavity	embryo	endosperm	embryo cavity	embryo cavity	full	infested	empty
Weight of 1,000 seeds	0.779	0.780	0.781	0.369	0.374	0.370	0.262	0.562	-0.400	-0.446
Length	0.780	0.776	0.776	0.387	0.441	0.307	0.270	0.514	-0.177	-0.551
Width	0.420	0.432	0.421	0.380	0.331	0.302	0.198	0.329	-0.088	-0.331
Volume	-0.074	-0.093	-0.104	0.067	0.000	0.008	0.080	0.116	-0.211	0.138
Area	0.665	0.692	0.685	0.452	0.455	0.365	0.253	0.416	-0.096	-0.478
light brown	0.138	0.209	0.198	-0.019	0.149	-0.018	-0.035	0.399	-0.257	-0.289
brown	-0.155	-0.165	-0.156	-0.007	-0.105	0.140	-0.140	-0.580	0.428	0.315
dark brown	-0.019	-0.055	-0.045	-0.096	-0.084	-0.060	-0.045	0.096	-0.121	0.028
cherry-red	0.049	-0.017	-0.024	0.080	-0.033	-0.173	0.294	0.270	-0.216	-0.093

Table 10. Coefficients of correlation between the seed internal structure characteristics and the characteristics of parent trees. Significant values (at $\alpha = 0.05$ level) are bold

Characteristics of parent trees (after BARAN 2005)	Weight of 1,000 seeds	Length			Width			Filling of embryo cavity		Seeds	
		embryo	endosperm	embryo cavity	embryo	endosperm	embryo cavity	embryo cavity	full	infested	empty
Height	0.097	0.088	0.046	0.032	-0.238	-0.102	-0.292	0.077	0.550	-0.557	-0.230
Crown length	0.011	-0.030	-0.051	-0.057	-0.260	-0.049	-0.277	-0.057	0.475	-0.458	-0.221
Relative crown length	-0.037	-0.085	-0.086	-0.086	-0.211	0.010	-0.201	-0.128	0.331	-0.299	-0.176
Crown width	0.020	-0.096	-0.060	-0.082	-0.152	-0.005	-0.096	-0.155	0.202	-0.267	0.022
dbh outside bark	0.071	-0.033	-0.040	-0.058	-0.209	-0.032	-0.166	-0.074	0.389	-0.467	-0.090
Bark thickness	0.207	0.100	0.110	0.081	-0.118	-0.028	-0.066	-0.085	0.500	-0.350	-0.312

of the investigated characteristics of the internal seed structure determined by the X-ray radiography. The weight of 1,000 seeds originating from all analyzed stands did not reach 55 g which is a long-term mean assumed for silver fir (ZAŁĘSKI, KANTOROWICZ 1993). The analysis of 1,000 seed weight has confirmed the intra-specific diversification of this characteristic observed during earlier studies (FOBER 1984; BODYŁ 2005) and showed the inter-population diversification which had no significant effect on seed germination capacity.

Silver fir provenances from Berest and Feleczyń in the Beskid Niski produced the heaviest seeds, while seeds of trees from Powroźnik and Kopciowa in the Beskid Sądecki were considerably lighter.

The percentage of seeds able to germinate was quite similar in all investigated stands, i.e. 43.5% on average. However, the percentage of viable seeds in the Kopciowa stand in the Beskid Sądecki (15.8%) was considerably different from the remaining stands. Possibly the lower viability of seeds in this stand resulted from a general weakening of this population observed during the year of seed collection.

The morphological and internal seed structure characteristics were found not to be significantly correlated with the growth parameters and crown structure of parent trees (BARAN 2005). It was interesting that seeds of plus trees in Powroźnik were characterized by lower values of the analyzed characteristics than were the mean values for seeds from the entire Powroźnik stand.

The seed wing colour was clearly associated with the occurrence of empty, full and insect damaged seeds. Seeds with brown wings were either empty or infested by insects to a high degree. In the Kopciowa Forest Range brown seed wings occurred on 81% of seeds, while the total proportion of empty and insect-infested seeds was 88%. The presence of the cherry-red colour was observed in a high proportion of full seeds able to germinate. These results have confirmed the considerable intra-specific variation of silver fir in the Carpathians (GUNIA 1986; SKRZYSZEWSKA 1999).

The results of this study showed that it is possible to widen and improve the methods of estimation of silver fir seed viability by taking into account the internal structure of seeds, including the embryo, endosperm, and filling of the embryo cavity. These characteristics, describing the potential germination capacity on the basis of the degree of embryo maturity, may constitute a convenient method of seed selection (separation) prior to the sowing operation, thus securing successful seed germination and good quality of planting stock produced.

The production of silver fir planting stock from qualified seed is still of basic importance in forestry. However, it makes forest management more difficult. Seeds are collected from standing trees, often over 40 m in height. Good seed year occurs rarely, every 3–4 years on average, and in mountains even every 5–8 years (ZAŁĘSKI, KANTOROWICZ 1993). In order to ensure the continuous planting stock production, also during poor seed years, and to protect endangered valuable silver fir populations in form of gene resources, it is necessary to store seed over periods of several years. Poor viability of silver fir seeds, in comparison with other coniferous species, causes that the forest nursery area sown with the low quality seed is quite considerable in size.

The silvicultural practice formed an opinion that heavier and larger seeds produce better seedlings. This should be explained by the better supply of the embryo with reserve materials during its maturation in the parent organism.

Studies of SABOR (1984) confirmed a significant interaction between the weight and the germination capacity of silver fir seeds. He found that the weight of seeds significantly affects their germination capacity. The probability of seed germination increases with an increase in seed weight irrespective of its moisture. Thus, germination success depends on medium-heavy and heavy seeds. However, it sometimes happens that heavy seeds, fully filled with the endosperm, have the embryo not fully developed or dead (ZAŁĘSKI 1995). In such a case the seeds, in spite of high weight, will not germinate after sowing.

This study has confirmed the usefulness of the X-ray radiography for the estimation of seed quality, mainly their viability and soundness. The comparison of these results with results of other authors is difficult due to the fact that most analyses carried out using this method concerned boreal tree species having seeds of high germination capacity conditioned by physiology, i.e. Scots pine and Norway spruce. Publications in this respect also concern tropical species. While e.g. KAMRA (1972, 1976) and FEDORKOV (2001) confirmed the usefulness of this method for the optimization of seed storage and nursery technology under European conditions, the tropical studies showed a limited use of the method. Therefore, the results presented in this paper, concerning the use of X-ray radiography for the seed quality estimation of silver fir, a species with rarely occurring seed years and producing seeds of low germination capacity (only 60% in the case of seed quality class 1), are of a significant economic importance in forestry in areas where silver fir is a stand-forming species.

Using X-ray photographs, showing the state of development of the embryo and endosperm, it is possible to improve the method of seed quality estimation, so important in seed storage and production of planting material. KAMRA (1972, 1976) confirmed the usefulness of X-ray radiography for the estimation of seed viability. He showed that this method is more reliable than the indigo-carmin staining for expeditious determination of germination capacity of Scots pine and Norway spruce seeds. He also considered this method as useful in determination of seed soundness, also in the case of tropical species. The use of X-rays in forest seed production has also been confirmed by Norwegian studies (BYE, EDVARDSEN 2004) indicating the possible use of this method for determination of viability of seeds in cones on the basis of the X-ray photographs showing the seed morphological development as well as the kind and extent of damage caused by insects and fungi. The studies of LEADEN et al. (1977) also confirmed the usefulness of X-ray photographs in determination of proportions of empty and full seeds as well as those damaged by insects in seed samples.

FEDORKOV (2001) confirmed the possible use of the X-ray method in estimation of seed quality of various provenances of Norway spruce and Scots pine when their transfer to different climatic zones is taken into consideration.

Taking into account the high costs of cone collection and seed extraction, the storage of low-quality seed would be a very uneconomical operation. The estimation of seed viability using their X-ray photographs, as described for example by ZAŁĘSKI et al. (2000), should take into account the following classification of the embryo and endosperm development: class I – seeds without embryos, class II – seeds with embryos taking up max. 50% of the embryo cavity length, class III – seeds with embryos taking up 51–75% of the embryo cavity length, classes IV and V – seeds with embryos taking up more than 75% of the embryo cavity length.

The results of the study presented in this paper showed that the estimation of the variation of seed morphological structure, significantly correlating with the variation of seed viability, makes the improvement of silver fir reproduction material in forest nurseries possible. Based on the seed wing colour, it also helps to determine the best date for the collection of physiologically mature and viable seeds.

CONCLUSIONS

1. This study has confirmed the possibility of using the X-ray method for expeditious verification of seed quality in silver fir (*Abies alba* Mill.). Its re-

sults have significantly complemented the present knowledge of the use of this method in reliable estimation of seed viability of tree species producing seeds of low germination capacity.

2. In the case of silver fir seeds, there is a significant effect of the stand (provenance) on the size of the embryo, endosperm, and embryo cavity. Also the provenance significantly affects the seed viability and proportions of seeds with underdeveloped embryo and endosperm as well as the degree of insect infestation of seeds.
3. The significant positive correlation of the degree of seed development, dimensions of the embryo and endosperm, as well as the length, width, surface area, and weight of seeds with the occurrence of full seeds, and the negative correlation of these characteristics with the occurrence of seeds infested by insects, as found during this study, may be utilized in elaboration of the methods of grading of silver fir seeds before they are sown in the nurseries, including container nurseries.
4. The analysis of development of the internal seed structure should be taken into account during the complex estimation of seed viability. Therefore, standards of mean sizes of the embryo and endosperm should be worked out for seeds of tree species having infrequently occurring good seed years and producing seeds of variable germination capacity. Such standards have already been elaborated for Scots pine and Norway spruce.
5. The relationship between the colour of seed wings and the germination capacity of seeds may help to determine the best seed collection date that would ensure the collection of high-quality silver fir seeds.

References

- BARAN P., 2005. Ocena wartości genetyczno-hodowlanej wybranych populacji jodły pospolitej (*Abies alba* Mill.) z terenu Leśnego Zakładu Doświadczalnego w Krynicy. [Praca magisterska.] Kraków, Katedra Nasiennictwa, Szkółkarstwa i Selekcji Drzew Leśnych. AR Kraków: 88.
- BELCHER E.W., 1974. Radiography in tree seed analysis has new twist. *Tree Planters Notes*, 24: 1–5.
- BODYŁ M., 2005. Zmienność żywotności i ciężaru nasion jodły pospolitej (*Abies alba* Mill.) w Polsce w zależności od regionu pochodzenia. [Maszynopis.] Warszawa, Instytut Badawczy Leśnictwa: 14.
- BYE H.R., EDVARDSEN Ø.M., 2004. The use of x-ray for prediction of seed viability in cones. Poster summary for the 27. ISTA Congress in Budapest. Available at: http://www.skogfroverket.no/Web/Informasjon/Artikler/X-ray_summary.pdf

- FEDORKOV A., 2001. Climatic adaptation of seed maturity in Scots pine and Norway spruce populations. *Silva Fennica*, 35: 119–123.
- FOBER H., 1984. Doświadczenie proveniencyjne nad jodłą pospolitą (*Abies alba* Mill.) założone w 1977 roku. *Arboretum Kórnickie*, 28: 145–157.
- GOZDALIK M., 1999. Zależność między wielkością zarodka i prąbielnią a wzrostem jednorocznych siewek sosny zwyczajnej (*Pinus silvestris* L.) i świerka pospolitego (*Picea abies* Karst.). [Ph.D. Thesis.] Warszawa, Forest Research Institute: 98.
- GUNIA S., 1986. Próba oceny wartości genetycznej i hodowlanej jodły pospolitej (*Abies alba* Mill.) z Sudetów i Karpat Polskich. *Sylvan*, 130: 83–92.
- KAMRA S.K., 1972. Comparative studies on germinability of *Pinus silvestris* and *Picea abies* seed by the indigo carmine and x-ray contrast methods. *Studia Forestalia Suecica*, 99: 1–21.
- KAMRA S.K., 1976. Use of X-ray radiography for studying seed quality in tropical forestry. *Studia Forestalia Suecica*, 131: 1–34.
- KAPUŚCIŃSKI S., 1966. Szkodniki owadzie nasion drzew leśnych. Warszawa, PWRiL: 156.
- LAFFERS A., 1979. Evaluation of seed weight in fir of Czechoslovakian and foreign provenance in relation to modified geographical latitude, to geographical longitude and to particular mountain ranges of Europe. *Lesnícky časopis*, 25: 111–125.
- LEADEM C.L., GILLIES S.L., YEARSLEY H.K., SIT V., SPITTLEHOSE D.L., BURTON P.J., 1997. Field Studies of Seed Biology. Land Management Handbook, No. 40, British Columbia, Ministry of Forests: 196.
- MACHANIČEK J., 1981. Výzkum kritérií určujících vhodnost lesního osiva pro dlouhodobé skladování. *Práce VÚLHM*, 59: 49–64.
- SABOR J., 1984. Zależność między ciężarem a zdolnością kiełkowania nasion jodły pospolitej. *Sylvan*, 127: 59–69.
- SALAZAR R., 1986. Seed and seedling provenance variation under greenhouse conditions of *Pinus caribaea* var. *hondurensis* Barr et Golf. *IPEF*, No. 32: 25–32.
- SIMAK M., 1974. Vaporous x-ray contrast agents for testing seed viability of *Pinus silvestris* L. In: *Proceedings: Seed X-ray Symposium*. Macon, USA: 29–39.
- SIMAK M., GUSTAFSSON A., 1954. Seed properties in mother trees and grafts of Scots pine. *Meddelanden från Statens Skogs-forskningsinstitut*, 44: 1–73.
- SIMAK M., 1980. X-radiography in research and testing of forest tree seeds. *Rapporter. Institutionen for Skogsskotsel, Sveriges Lantbruksuniversitet*, 3: 34.
- SKRZYSZEWSKA K., 1999. Wartość genetyczno-hodowlana jodły pospolitej (*Abies alba* Mill.) reprezentowanej w Ogólnopolskim Doświadczeniu Proveniencyjnym Jd PL 86/90. *Zeszyty Naukowe AR w Krakowie*, 339, Seria Sesja Naukowa, 61: 43–66.
- SKRZYSZEWSKA K., 2007. Genetic structure of silver fir (*Abies alba* Mill.) in Sącz Beskid. *Beskydy*, 20: 243–250.
- SMIRNOVA N.G., 1978. Rentgenografičeskoje izučeniye siemjan listviennych dreviesnykh rastenij. Moskva, Akademiya Nauk SSSR, Izdatel'stvo Nauka: 243.
- Statistica ELECTRONIC MANUAL (w:) StatSoft, Inc. (2001). STATISTICA (data analysis software system), Version 6. www.statsoft.com. wersja polska.
- ZAŁĘSKI A., 1990. Wpływ wielkości zarodka w nasionach gatunków iglastych na udatność siewów i rozwój siewek. *Biuletyn Instytutu Badawczego Leśnictwa*, 14 (2): 15–17.
- ZAŁĘSKI A., 1995. Rentgenowskie metody oceny żywotności nasion. In: ZAŁĘSKI A. (ed.), *Nasiennictwo leśnych drzew i krzewów iglastych*. Warszawa, Wydawnictwo Świat: 130–134.
- ZAŁĘSKI A., BORKOWSKA E., 1993. Zależności między wielkością zarodka i bielnią a wzrostem jednorocznych siewek sosny czarnej (*Pinus nigra* Arn.). *Prace Instytutu Badawczego Leśnictwa, Seria A*, 752: 3–17.
- ZAŁĘSKI A., KANTOROWICZ W., 1993. Obrządzanie najważniejszych gatunków drzew leśnych w latach 1957–1991. *Notatnik Naukowy Instytutu Badawczego Leśnictwa*, 2: 1–7.
- ZAŁĘSKI A., ANIŚKO A., KANTOROWICZ W., SOB-CZAK H., 2000. *Zasady i metodyka oceny nasion w Lasach Państwowych*. Warszawa, CILP.

Received for publication May 28, 2008

Accepted after corrections April 10, 2009

Studie variability morfologicznych charakteristik semen jedle białokoré (*Abies alba* Mill.) a jejich vnitřní struktury určených rentgenovou radiografií v horských pásmech Beskid Sądecki a Beskid Niski v Karpatech (jižní Polsko)

ABSTRAKT: Byla hodnocena variabilita morfologicznych charakteristik a vnitřní struktury semen jedle białokoré (*Abies alba* Mill.) ze čtyř stanovišť v nadmořské výšce mezi 625–750 m ve dvou horských pásmech Karpat v jižním Polsku – Beskid Sądecki a Beskid Niski. Sběr semen proběhl ve druhé polovině září 2004. Byla určena hmotnost

1 000 vzduchem sušených semen a jejich životaschopnost byla hodnocena pomocí rentgenové radiografie. Délka a šířka embrya, endospermu a embryonální dutiny byly měřeny na rentgenových snímcích. Také morfologické znaky semen, tj. délka, šířka, objem, povrchová plocha a barva křídélek (světle hnědá, hnědá, tmavě hnědá, třeshňově červená), byly zahrnuty do této analýzy. Byl zjištěn statisticky významný vliv provenience na zkoumané parametry embrya, endospermu a embryonální dutiny i na výskyt plných a hmyzem napadených semen. Hmotnost 1 000 semen byla nižší než 55 g, což je považováno za dlouhodobý průměr pro jedli bělokrou v Polsku. Byla nalezena pozitivní korelace hmotnosti semen s délkou a šířkou embrya, endospermu a embryonální dutiny a stejně tak i se stupněm vyplnění embryonální dutiny embryem. Hmotnost semen rovněž pozitivně korelovala s jejich délkou, šířkou a povrchovou plochou. Byla zjištěna i korelace mezi barvou křídélek a hmotností 1 000 semen či poměrem hmyzem napadených a prázdných semen. Podíl semen s hnědými křídélky negativně koreloval s hmotností 1 000 semen a počtem plných semen, zatímco pozitivní korelace byla zjištěna ve vztahu s počtem semen napadených hmyzem a prázdných semen. Výskyt světle hnědých a třeshňově červených křídélek měl souvislost s velkým podílem plných semen. Výsledky korelace mezi parametry semen a vybranými charakteristikami testovaných stromů indikují statisticky významné vztahy. Pozitivní korelace byla zjištěna mezi podílem plných semen a výškou stromu, tvarem koruny, průměrem v prsní výšce a tloušťkou kůry. Oproti tomu byl zjištěn negativní vztah mezi těmito charakteristikami stromů a podílem hmyzem napadených semen.

Klíčová slova: *Abies alba*; X-paprsek; životaschopnost semene; vnitřní struktura semene; morfologické charakteristiky semene; Karpaty

Corresponding author:

Dr. inż. KINGA SKRZYSZEWSKA, Agricultural University of Cracow, Faculty of Forestry, Department of Forest Trees Breeding, Al. 29 Listopada 46, 31-425 Cracow, Poland
tel.: + 48 12 662 51 26, fax: + 48 12 662 51 28, e-mail: rlkskrzy@cyf-kr.edu.pl
