Genetic diversity and spatial structure of the selected young Norway spruce (*Picea abies*) population in the Giant Mts.

Genetická diversita a prostorová struktura vybrané mladé populace smrku (*Picea abies*) v Krkonoších

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Abstract
Genetic diversity and spatial structure of a young Norway spruce (*Picea abies* Karst.) stand was investigated within the planted stand localized in the Giant Mts. near Špindlerův Mlýn. Average tree height was 15.1 m, average diameter at breast height was 245 mm. The spatial distribution of trees was random. Spruce branches of all trees in selected part of the stand were sampled in order to accomplish isozyme analysis. One-dimensional horizontal electrophoresis on starch gel was used whereas present alleles of isozyme loci G-6-PDH, GDH, SDH-A, SDH-B PGM-A, IDH-B, AAT-B and AAT-C were determined. Subsequently, allele numbers per loci, allelic frequency and heterozygosity were evaluated. Genotype combinations were a basis to calculate coefficients of genetic diversity: Shannon's index and Rao's quadratic entropy. Genotypes were not randomly distributed in the stand. Trees with some alleles are clumped. Further, three variants of thinning was simulated: Total 30 % of trees would be cut and 30 % represents trees with the highest diameter (variant v1), 15 % represents trees with the highest diameter and 15 % represents trees with the lowest one (v2), all 30 % are trees of the lowest diameter (v3). The genetic diversity is affected with the simulated thinning.

Abstrakt
Byla sledována genetická diversita a prostorová struktura mladého vysázeného porostu smrku (*Picea abies* Karst.) v Krkonoších poblíž Špindlerova Mlýnu. Průměrná výška stromů byla 15,1 m a výčetní tloušťka 245 mm. Prostorové rozmístění stromů bylo náhodné. V části porostu byly odebrány vzorky větví všech jedinců smrku pro získání materiálu pro isoenzymové analýzy. Pro tyto vzorky byla užita jednorozměrná horizontální elektroforéza na škrobovém gelu. Sledovány byly lokusy G-6-PDH, GDH, SDH-A, SDH-B PGM-A, IDH-B, AAT-B a AAT-C. Byl vyhodnocen počet alel na lokus, alelické frekvence a hodnoty heterozygotnosti. Základem pro výpočet indexů diversity (Shannonův index a Rao kvadratická entropie) byly genotypové kombinace. Jednotlivé genotypy nebyly rozmístěny v porostu náhodně. Následně byly simulovány tři varianty probírky s odstraněním celkem 30 % stromů: 30 % representuje stromy s největším průměrem (varianta v1), 15 % stromů s největším průměrem a 15 % stromů s nejnižším průměrem je odstraňováno (v2), 30 % představuje stromy s nejnižším průměrem (v3). Simulovanou probírkou byla ovlivněna genetická diversita porostu.

Keywords: genetic diversity, heterozygosity, isozyme, *Picea abies*, population structure, spatial structure
Klíčová slova: genetická diversita, heterozygotnost, isoenzymy, *Picea abies*, populační struktura, prostorová struktura

Introduction

The Norway spruce (*Picea abies* (L.) Karst.) dominated ecosystems represent a typical kind of forests near the alpine timberline in the Central Europe (SCHMIDT-VOGT 1977). These ecosystems grow in extreme environmental conditions. The spruce forests in the Czech Republic were often under high air-pollution impact, which has caused dieback of the stands. Such situation was observed in the Giant Mountains (Krkonoše Mts.) in a large area (VACEK et al. 2007). Wind is another environmental factor has caused the large-area disintegration of the tree stands during recent history in the Giant Mountains. These facts lead to existence of many ecosystems with young Norway spruce stands in sites near alpine timberline (8th forest altitudinal zone in sense of the Czech forest typological system; VIEWEGH et al. 2003). Under such circumstances, the studying of genetic properties of these young populations and possibilities of their further development is of outstanding importance.
Knowledge on the Norway spruce genetic features has the wide background in the literature.
Many studies in the forest genetics have used isozymes belonging to the common types of molecular gene markers. Isozymes are various forms of an enzyme or other active soluble protein with different electrophoretic mobilities which act on a given substrate (PASTEUR et al. 1988). The isozyme analysis represents a method useful to description basic genetic variation both within a population and between different populations.

Geographical isozyme variability

The large-area oriented research of the Norway spruce isozyme variation in Europe was carried out in several projects (KANNENBERG & GROSS 1999; PERRY et al. 1999; KRUTOVSKII & BERGMANN 1995). Isozyme polymorphism of the Norway spruce populations from 15 European provenances, which had been included into the IUFRO testing programme, showed that some alleles of the SDH-A, IDH-A, LAP-A and GDH loci exhibited a geographic pattern. The measures of genetic diversity showed lower values in Southern and Central Europe than in northern and north-eastern part of the continent, probably resulting from introgressive hybridization with other species (KRUTOVSKII & BERGMANN 1995). For most provenances, significant correlations were found between genetic dissimilarity and geographic distance (KANNENBERG & GROSS 1999). The genetic diversity and differentiation was also described in the Italian spruce populations by GIANNINI et al. (1991) who found that only small part of the total genetic variation is due to interpopulational differentiation. On the other hand, no variation of allele frequency patterns along latitude or altitude in Norway spruce populations was found in the small geographical region - Black Forest (Schwarzwald) (KONNERT 1991).

Regional studies of isozyme systems by Norway spruce are known from Italy (GIANNINI et al. 1991), Austria (GEBUREK 1998), Germany (KONNERT 1991a, b, KONNERT & FRANKE 1991), Poland (BURCZYK et al 2004; CHAŁUPKA et al. 2008), Latvia (GONCHARENKO et al. 1995) and Ukraine (KORSHIKOV et al. 2008). The study of spruce seed orchards (PAULE et al. 1993) belongs to the first isozyme studies of the Norway spruce populations, stands and seed orchards in region of the former Czechoslovakia. Norway spruce populations from the Ore Mts. and Giant Mts. were compared on the base of isozyme analyses in framework of different environmental conditions (IVANEK 2000). MÁNEK (1999, 2001) was concerned in the Norway spruce stands in the Šumava Mts., having compared genetic characteristics of three autochthonous populations. Genetic study of the Norway spruce populations Czech Republic was accomplished for 10 ICP-Forests monitoring plots. The results suggested that genetic diversity of the Norway spruce populations was determined both by reproductive material used and by genotype selection under differentiated stand factors (IVANEK 2006).

Population spatial pattern

Natural populations of plants can be spatially organized. This spatial pattern can be observable according to the genetic features, too. There are known statistical procedures which are suitable to reveal these relations (e.g. HARDY & VEKEMANS 2002).

Using isozymes as gene markers and spatial autocorrelation of allozyme traits, random distribution of genotypes in space was found in most cases in the model spruce populations from the eastern Italian Alps. However a few genotypes showed a significant clumped distribution over a small spatial scale (LEONARDI et al. 1996).

Weak spatial pattern and non-random association of genotypes was observed within autochthonous, high-elevation Norway spruce in the Austria what was explained as a result of microselection or statistical artefact (GEBUREK 1998).

Research in the Giant Mountains

The isozyme analysis of dormant buds of the Norway spruce clonal plantations from nurseries in the Giant Mts. was performed in order to certificate clonal homogeneity. Molecular genetic characteristics of the clones were compared with growth and phenology characteristics (IVANEK & MARTINCOVÁ 2005, 2006). Genetic differences between two Norway spruce tree subpopulations in the Giant Mountains (Krkonoše in Czech) National Park, the Sněžka Mt. region, differing markedly in extent of damage after the Kyrill hurricane (2007), were investigated using isozyme analysis and biometric measurements including preliminary tree-ring analysis. Genetic distances of investigated spruce sets in framework of overall six spruce populations were calculated (IVANEK et al. 2009).

Aim of this study can be stated in the following points:

- description of genetic structure of the tree population using selected isozyme markers;
- study relationships of genetic structure to spatial structure;
- to give evidence on differences among phenotype groups of the young spruce;
- to assess possibility for a genetic structure change on the base of different models of intervention - thinning.

A young Norway spruce planted stand was selected as the model population. Several first results were published by IVANEK & MATĚJKA (2009).

Methods

Stand description and sampling

The young Norway spruce stand was investigated within the planted stand (age of 39 years at 2008) localized in the Giant Mts. near Špindlerův Mlýn, forest enterprise Vrchlabí, stand group $202F_4$, altitude 1080 m, inclination 25°, south-western orientation.

Actual vegetation can be described by the relevé (number 131/09, date 25.8.2009): total cover E_3 70 %, E_2 0 %, E_1 25 %, E_0 60 %. E_3: *Picea abies* 4, *Sorbus aucuparia* +; E_1: *Avenella flexuosa* +, *Blechnum spicant* r, *Calamagrostis villosa* 2, *Carex echinata* r, *Carex ovalis* +, *Dryopteris dilatata* 1-2, *Galium saxatile* +, *Gentiana asclepiadea* 1, *Hieracium lachenalii* +, *Homogyne alpina* +-1, *Juncus effusus* r-+, *Oxalis acetosella* 1, *Polygonatum verticillatum* r, *Rubus idaeus* +, *Rumex arifolius* r, *Senecio ovatus* 1-2, *Sorbus aucuparia* r, *Vaccinium myrtillus* +. This community corresponds to a transition between 7th and 8th forest altitudinal zone.
The research plot is represented by a continuous area where all Norway spruce trees have been sampled. Spruce branches of all trees in selected part of the stand were sampled (spring 2008) in order to accomplish isozyme analysis. Each tree was permanently marked at this time. All samples were transported into the laboratory immediately. The collected samples were stored in the freezer storage box by the time of analyze.
Position of each tree was measured using the Field-Map technology (www.fieldmap.cz) at May 2009. Tree perimeter at breast height (which was recalculated to tree diameter - DBH) and tree height (h) was recorded as the basic tree size features. The crown defoliation was assessed in per-cents. Occurrence of the trunk damage (breaks, forks, trunk curvature, visible fungus infection etc.) was recorded. The spruce individuals were separated in classes according to branching using known system (SAMEK 1964; SCHMIDT-VOGT 1972), which was simplified regarding to the young population without sufficient manifestation of attributes to be used.
The plot limit trees were selected in GIS. Distances of the tree to several first neighbourhood trees were calculated. Such distance to the first closest neighbour is marked as d_1, similar distance to the second closest neighbour is marked as d_2 and so forth. The plot border was constructed as the outer buffer around the line connecting the limit trees. The buffer width was equal to one half of arithmetic mean of d_2 for all not-limit trees.

Isozyme analysis

Dormant buds from sampled branches were collected and homogenized with modified extraction buffer (pH 6.7) for tissues with high levels of interfering substances (WENDEL & WEEDEN 1990). The isozymes were separated by horizontal one-dimensional electrophoresis of the homogenate on starch gel at 3 - 5 °C in the Tris-citrate buffer (pH 7.5) using Multiphor II electrophoretic device. Each sample was randomly located at two gels. Each gel was completed by one comparative standard sample. Eight isozyme systems (Table 1) were stained according to PASTEUR et al. (1988) at 37 °C. The gels were scanned and evaluated with ImageMaster software (Pharmacia Biotech).

Data processing

The spatial data was processed in GIS (TopoL software; www.topol.cz) and using special software (PlotOA; www.infodatasys.cz/software).
Data exported from the ImageMaster programme was stored into the IsoEnz database (MATĚJKA 2009a). Each allele pair determination was carried out on the base of relative mobility using the SeqAn programme (MATĚJKA 2009b). The comparison within set of all gels was used in order to a unique identification of alleles over all trees analysed in a few gels. Alleles were numbered in increasing order.
Allele numbers per loci, allelic frequency and observed heterozygosity were evaluated as measures related to the single locus. Differences between observed heterozygosities h_1 and h_2 in the populations 1 and 2 of the population sizes n_1 and n_2 was statistically tested using test variable

$$t = \frac{h_1 - h_2}{\sqrt{\left(h_1(1-h_1)/n_1 - 1\right) + \left(h_2(1-h_2)/n_2 - 1\right)}}$$

which has the Student t-distribution with n_1+n_2-2 degrees of freedom (e.g. ŠKRÁŠEK & TICHÝ 1990: pp. 259-262).
Indices of diversity were evaluated using all processed enzymatic systems. Each individual tree was assigned in a genotype class using the combination of all studied allele pairs. For instance, it is possible to use the sequence "222223222222222212" as an identification string of the genotype class identified on the base of the sequence of G6PDH (22), GDH (22), SDH-A (23), PGM-A (22), MDH-B (22), MDH-C (22), IDH-B (22), AAT-B (22) and AAT-C (12) enzyme loci. Genetic diversity of the population was evaluated in the following manners.
Number of classes over combinations of all-loci (number of different identification strings) is the simplest parameter.
Potential number of classes (c_{pot}) - theoretical number of all possible identification strings combining all alleles presented in the investigated set of trees can be viewed as indicator of diversity in a theoretical population of unlimited size.

The Shannon's index $H = -\sum_{i=1}^{c} p_i \ln_2 p_i$ (where c is number of the classes; p_i is relative share of trees within the i-th class; SHANNON & WEAVER 1949) was modified in the population genetics in several cases earlier (e.g.

AFIF et al. 2008; LIA et al. 2008). Equitability $e = H / \ln_2 c_{pot}$ completes set of the indices. The last index is the Rao coefficient $R = \sum_{i=1}^{c} \sum_{j=1}^{c} d_{ij} p_i p_j$, d_{ij} has been calculated according to the equation $d_{ij} = a_{ij} / 2N$, where a_{ij} is number alleles to be not contained in both identification strings and N is number of loci. This index is an adoption of the quadratic entropy measure (RAO, 1982) in population genetics. All indices were calculated using the IsoEnz software (MATĚJKA 2009a).

The population was divided into two or more subpopulations according to a selected feature. Dissimilarity between subpopulations was calculated according to NEI (1978).

Results and discussion

The stand structure

The 112 trees were selected (108 individuals of Norway spruce and 4 rowan polycormons). Total 36 trees were indicated as limit. The average tree-to- neighbour distances were d_1 = 1.9 m, d_2 = 2.8 and d_3 = 3.6. Total plot area was 1391 m^2. The Norway spruce density was 777 individuals per 1 ha.

Average tree height was 15.1 m (standard deviation ±2.3 m; total 106 individuals), average diameter at breast height was 245 ± 50 mm (2009). Mean tree defoliation was 29 ± 15 % with interval 5 - 90 %. One quarter of trees was with damaged trunks. Average parameters of undamaged trees (n = 78) were slightly different: tree height 15.8 ± 1.3 m, DBH 258 ± 42 mm and defoliation 24 ± 9 %.

Nevertheless more branching types are known by Norway spruce (SAMEK 1964; SCHMIDT-VOGT H. 1972), only two approximate classes were possible to distinguish in the young population. Trees with the plate-shape branching prevail. The class of comb-shape types includes wide variability from brush- to comb-shaped branching. The trunk damage differs in both plate- and comb-shape types significantly (χ^2-test, p = 0.006; Table 2A). The two-factor ANOVA reveals that trunk damage had an effect on all variables - tree height, DBH and defoliation. Tree membership to the group according to branching type had no effect although average tree-size variables were slightly higher in the comb-shape type group and average defoliation was lower (Table 2).

Genetic structure of the population

Polymorphism was found in all investigated loci except IDH-A whereas high allele number was observed by AAT-C and SDH-A. Rare allele of AAT-B and very rare allele of SDH-A were found (Table 3). This is comparable with the Norway spruce polymorphism of these loci in the Central Europe (KONNERT & MAURER 1995). Observed polymorphism was higher in comparison with 11 planted plots studied within the ICP-Forests programme (IVANEK 2006). The data collected in the Bohemian Forest (MÁNEK 1999) are specific regarding to allele numbers for the SDH-A and LAP-B loci in particular. Comparing coefficients H and R in the studied population (Table 5) with several other spruce stands in the Czech Republic (IVANEK et al. 2009), moderate genetic diversity of the investigated spruce stand can be claimed. Lower value of overall allele number but increased number of classes for the same set of isozyme loci were found in comparison with the mature Norway spruce population growing near Sněžka Mt.

Spatial structure

Spatial distribution of trees in the stand in relation to the tree diameter at breast height is shown in the Fig.1.

Genotypes are not randomly distributed in the stand. Trees with some alleles are clumped. Such typical example could be demonstrated by AAT-C and SDH-A (Fig. 2, 3) what is in accordance with clumped distribution over a small spatial scale reported by (LEONARDI et al. 1996). Considering relatively low stand age, the microselection (GEBUREK 1998) can be excluded.

Phenotype differentiation

Comparing genetic diversity of two phenotype groups, higher observed AAT-C and SDH-A heterozygosity was found in the group of comb-shape phenotypes. Difference between the G-6-PDH heterozygosities was statistically significant. On the other hand, substantial increase in number of classes and potential number of classes and moderate increase of genetic diversity coefficients (H, R) were found for the plate-shape types (Table 6). The Nei-distance between two subpopulations of both phenotype groups is 0.0036 - this value is comparable with the distance between populations growing in a region (Bohemian Forest, 0.004; MÁNEK 1999) and lower than the distance between populations of origin in different regions (Giant Mts. - Jeseníky Mts., 0.068; IVANEK et al. 2009).

Model thinning

Total 30 % of trees would be cut according to three simulated variants of thinning:

- 30 % represents trees with the highest diameter (v1),

- 15 % represents trees with the highest diameter and 15 % represents trees with the lowest one (v2),
- all 30 % are trees of the lowest diameter (v3).

Decrease of heterozygosity was observed in the variant v1 for the G-6-PDH, GDH, MDH-C and AAT-C loci comparing with other variants of the model thinning. Decrease of allele number per locus was observed in the variant v3. Further, heterozygosity loss was found for the MDH-B, MDH-C, and AAT-B loci here. The lowest heterozygosity was found in the variant v3. Statistical importance of the heterozygosity differences is minor (Table 4).

Number of genotype classes was reduced in all variants. Potential number of genotype classes was unchanged in the variant v1 and substantially reduced in the variant v3. The Shannon's index was the lowest in case of the v3 variant. The Rao's index was not significantly affected by thinning (Table 5).

The Nei distance among three hypothetical populations and the original population without thinning were calculated (Fig. 4). The highest distance (0.00024) was observed between variants v1 and v3. This value is four-time lower than dissimilarity between two mature Norway spruce subpopulations in the locality near Sněžka Mt. (IVANEK & MATĚJKA 2009).

Differentiation according to damage and defoliation

Trees with damaged trunks (including breaking, doubled trunk etc.) represent group of a different genetic constitution, but with the same amount of diversity comparing undamaged trees. The heterozygosity difference is near to significant by the AAT-C locus (Table 7). Differentiation of both subpopulations is important - the corresponding Nei-distance was 0.0054. There is no important difference between the whole set of trees and that one corresponding to the trees with defoliation ≤ 30% (Table 8). Thinning the most defoliated trees is probably without any important effect on genetic diversity.

Conclusions

Genetic structure of the planted Norway spruce population shows important spatial dependencies. Clumps of the similar genotypes show diameter in order of 10 m.

Subpopulation of the comb-shaped types of trees has different genetic composition comparing the other subpopulation.

Thinning models according to tree size point to the best thinning in favour to preserve genetic diversity – it is combined thinning (the variant v2).

Set of damaged trees (with breaking, doubled trunk, other damaged trunk) has different genetic composition comparing other trees.

Defoliation of young trees (about 40 years) is probably not fitting feature to separate tree set of different genetic structure.

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Table 1. Assessed enzymes and scored loci.

Enzyme	E. C. No.	Scored loci
Glucose-6-phosphate dehydrogenase	1. 1. 1. 49	G-6-PDH
Glutamate dehydrogenase	1. 4. 1. 2	GDH
Shikimate dehydrogenase	1. 1. 1. 25	SDH-A
Phosphoglucomutase	2. 7. 5. 1	PGM-A
Malate dehydrogenase	1. 6. 99. 3	MDH-B, MDH-C
Leucine aminopeptidase	3. 4. 11. 1	LAP-B
Isocitrate dehydrogenase	1. 1. 1. 42	IDH-A, IDH-B
Aspartate aminotransferase	2. 6. 1. 1	AAT-A, AAT-B, AAT-C

Table 2. Differentiation of basic dendrometric features between selected groups of trees.

A. Number of individuals.

	Damaged	Undamaged
Plate-shape types	57	27
Comb-shape types	21	1

B. Tree height - mean ± standard error (in m).

	Damaged	Undamaged
Plate-shape types	15.5±0.3	13.1±0.4
Comb-shape types	16.6±0.4	12.9±2.0

C. DBH - mean ± standard error (in mm).

	Damaged	Undamaged
Plate-shape types	250±6	213±9
Comb-shape types	278±10	140±44

D. Defoliation - mean ± standard error (in %).

	Damaged	Undamaged
Plate-shape types	26±2	42±2
Comb-shape types	19±3	40±12

Table 3. Number of alleles in the Norway spruce populations referred in some studies.
Null alleles were excluded. n.d. - not determined.

Data set	This study	Central Europe (KONNERT & MAURER 1995)	ICP Forests (IVANEK 2006)	Bohemian Forest (MÁNEK 1999)
AAT-B	2	3	n.d.	2
AAT-C	4	5	n.d	3
G-6-PDH	2	4	2	n.d.
GDH	2	3	2	2
IDH-A	1	6	1	3
IDH-B	2	3	1	2
LAP-B	2	10	n.d.	5
MDH-B	2	3	1	1
MDH-C	2	5	1	3
PGM-A	2	4	2	2
SDH-A	5	7	4	2

Table 4. Effect of thinning according to tree size on observed heterozygosity. Differences were evaluated by the t-test.

Set of trees	all		with low DBH	with middle DBH	with high DBH	Maximal difference and their probability (p)
Thinning variant	none		v1	v2	v3	
	Alleles	Observed heterozygosity				
AAT-B	2	0.0097	0.0141	0.0139	0	v1 - v3: 0.149
AAT-C	4	0.5208	0.4923	0.5672	0.5882	v1 - v3: 0.111
G-6-PDH	2	0.1226	0.0959	0.1233	0.1507	v1 - v3: 0.155
GDH	2	0.0196	0.0141	0.0278	0.0286	v1 - v3: 0.267
IDH-B	2	0.0094	0.0137	0.0137	0.0137	v1 - none: 0.397
MDH-B	2	0.0097	0.0141	0.0139	0	v1 - v3: 0.149
MDH-C	2	0.0485	0.0563	0.0694	0.0423	v2 - v3: 0.235
PGM-A	2	0.0094	0.0137	0	0	v1 - v3: 0.156
SDH-A	5	0.2547	0.2603	0.2740	0.2740	none - v3: 0.387

Table 5. Effect of thinning according to tree size on selected features of genetic diversity.

Set of trees	all	with low DBH	with middle DBH	with high DBH
Thinning variant	none	v1	v2	v3
Classes	31	25	27	25
Potential classes no.	328050	328050	109350	12150
H	4.1162	4.0872	4.2014	3.9504
e	0.2246	0.2231	0.251	0.2911
R	0.0975	0.0997	0.1031	0.0949

Table 6. Comparing genetic diversity of two phenotype groups. p - error probability by the t-test of heterozygosity difference.

Phenotype	Plate-shape types	Comb-shape types	p
Tree count	85	22	
Observed heterozygosity:			
AAT-B	0.013	0	0.148
AAT-C	0.493	0.650	0.083
G-6-PDH	0.145	0.046	0.049
GDH	0.013	0.048	0.228
IDH-B	0.012	0	0.157
MDH-B	0.013	0	0.148
MDH-C	0.050	0.046	0.465
PGM-A	0.012	0	0.157
SDH-A	0.241	0.318	0.246
Diversity:			
Number of classes	28	13	
Potential classes no.	196830	2700	
H	4.0434	3.4714	
e	0.2299	0.3045	
R	0.1006	0.0803	

Table 7. Genetic differentiation according to trunk damage. p - error probability by the t-test of heterozygosity difference.

	Damaged trees	Undamaged trees	p
Tree count	28	78	
Observed heterozygosity:			
AAT-B	0	0.013	0.151
AAT-C	0.333	0.592	0.006
G-6-PDH	0.107	0.130	0.374
GDH	0	0.027	0.068
IDH-B	0	0.013	0.355
MDH-B	0	0.013	0.151
MDH-C	0.037	0.053	0.355
PGM-A	0.036	0	0.160
SDH-A	0.179	0.286	0.118
Diversity:			
Number of classes	12	29	
Potential classes no.	972	109350	
H	3.1887	4.1478	
e	0.3213	0.2478	
R	0.0853	0.0985	

Table 8. Differentiation according to defoliation

	All trees	Trees with defoliation ≤ 30%
Tree count	106	79
Observed heterozygosity:		
AAT-B	0.010	0.013
AAT-C	0.521	0.595
G-6-PDH	0.123	0.114
GDH	0.020	0.026
IDH-B	0.009	0.013
MDH-B	0.010	0.013
MDH-C	0.049	0.051
PGM-A	0.009	0
SDH-A	0.255	0.253
Diversity:		
Classes	31	27
Potential classes no.	328050	109350
H	4.1162	3.9406
e	0.2246	0.2354
R	0.0975	0.0933

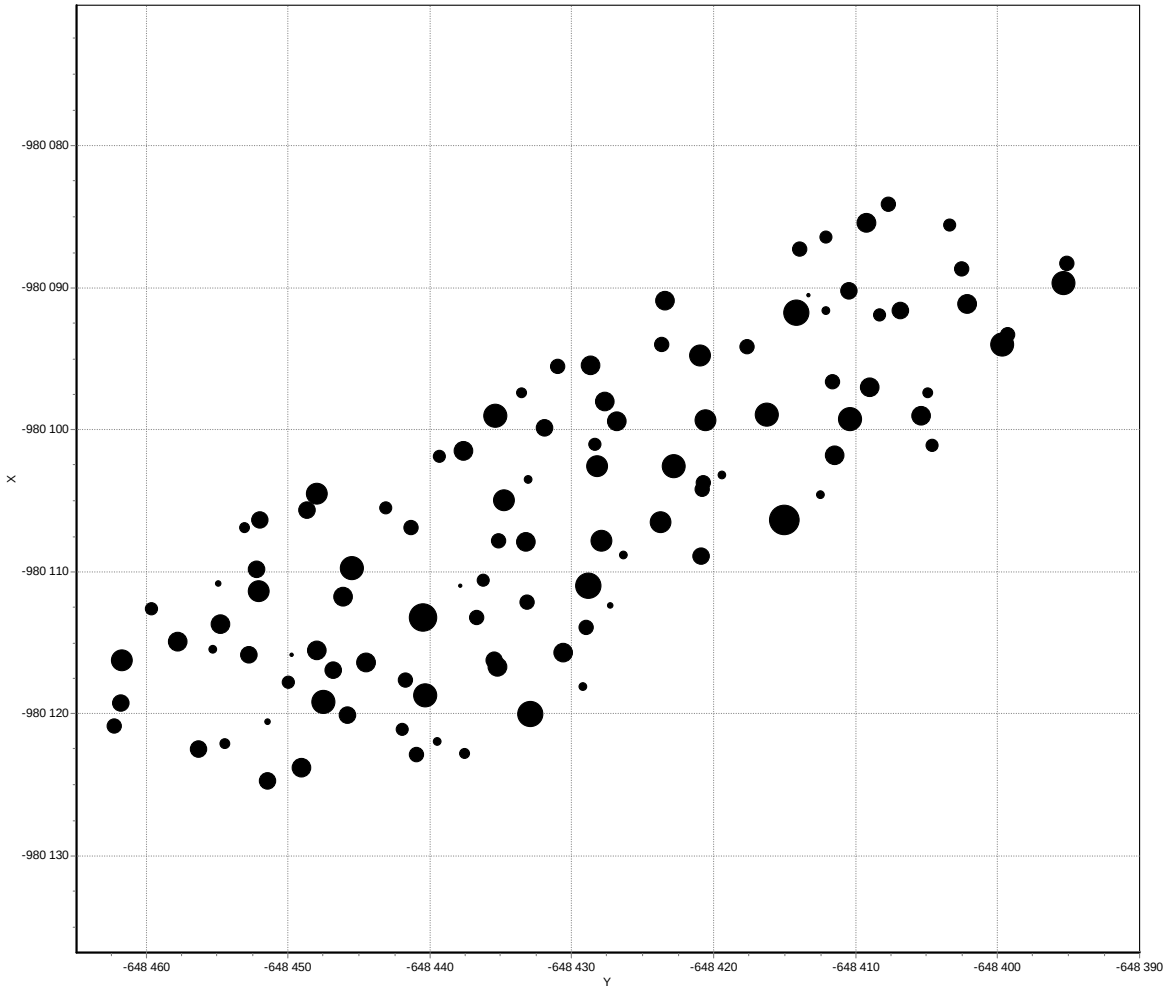

Fig. 1. Spatial distribution of trees in the stand. Point size is related to tree diameter at breast height.

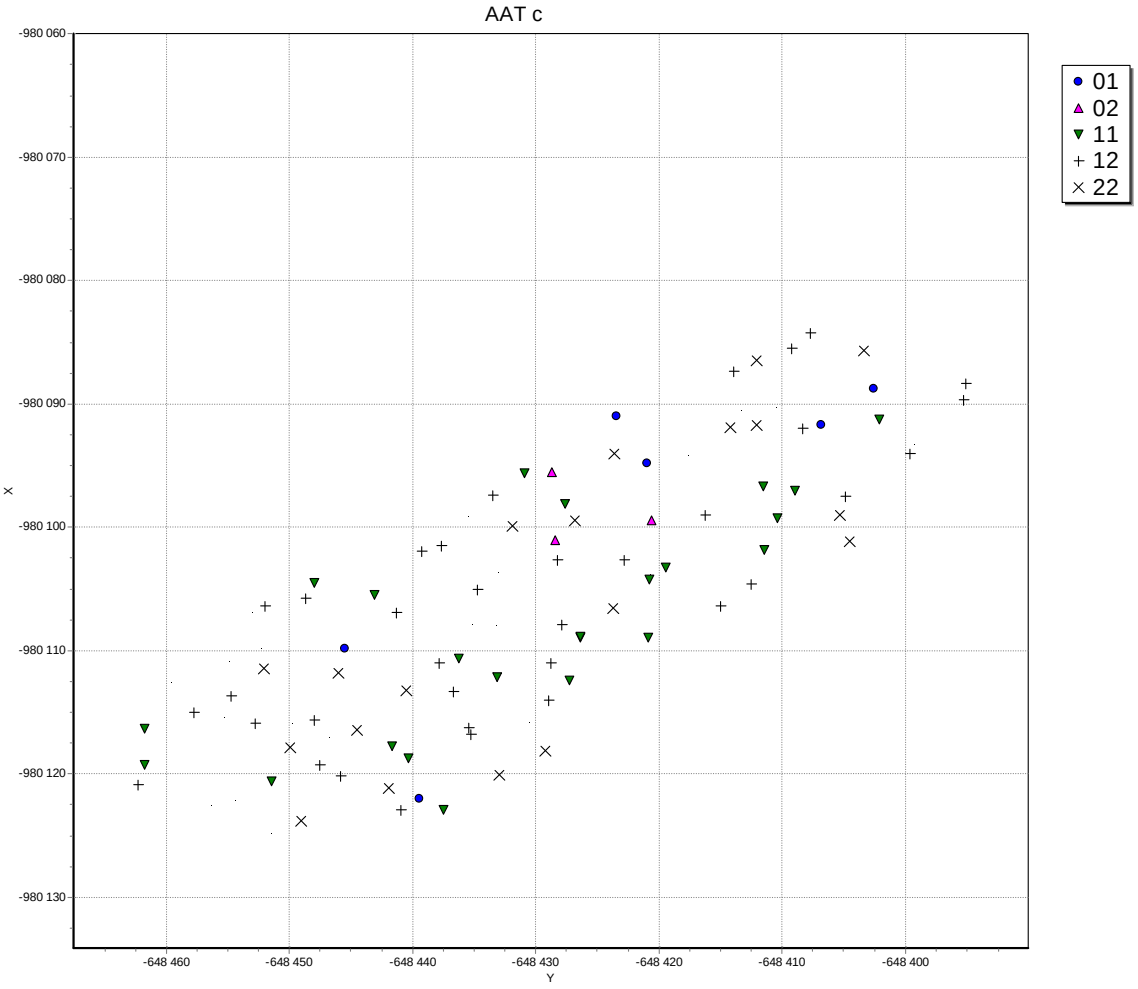

Fig. 2. Spatial distribution of trees in the stand according to AAT-C allele combinations.

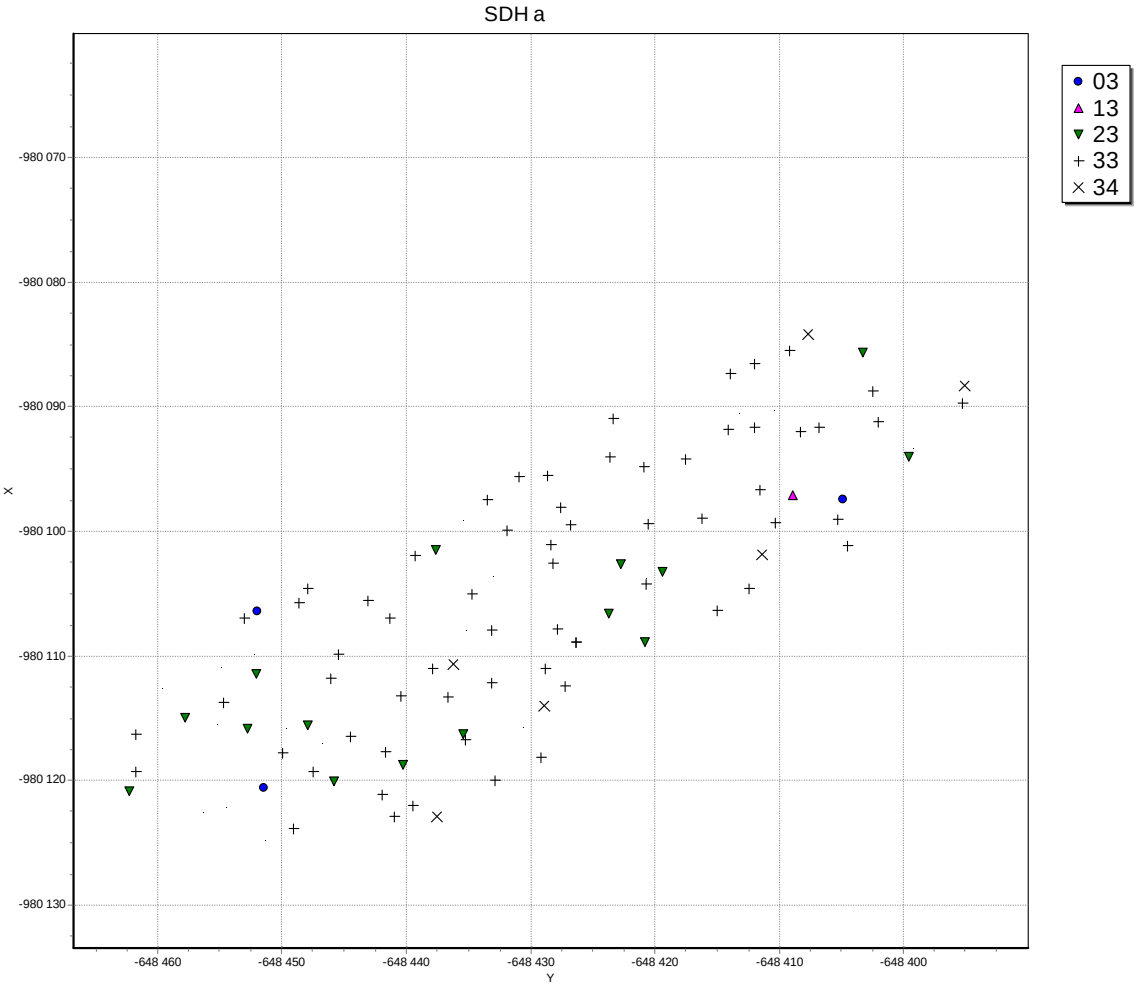

Fig. 3. Spatial distribution of trees in the stand according to SDH-A allele combinations.

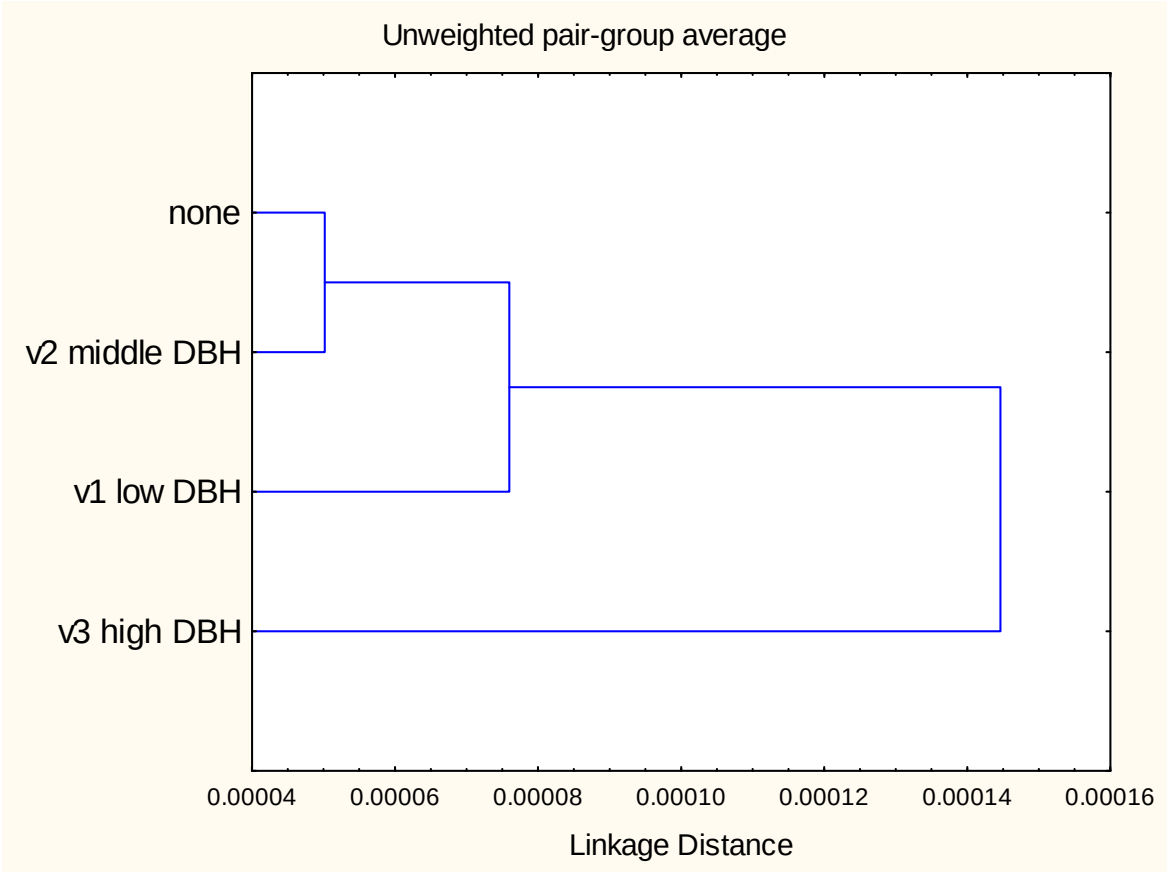

Fig. 4. Classification of the hypothetical Norway spruce populations formatted by different thinning (see Tables 3-4). The Nei distance was used.