

LEON MEJNARTOWICZ, STEFAN BIAŁOBOK, PIOTR KAROLEWSKI

Genetic characteristics of Scots pine specimens resistant and susceptible to SO₂ action*

Studies on the variability in the degree of damage to woody plants by gaseous emissions are of increasing interest to scientists dealing with nature conservancy.

The aim of presented work was to determine the relations between the degree of resistance of Scots pine to SO₂ action and the degree of heterozygosity of individuals in an indigenous population.

MATERIALS AND METHODS

The Scots pine individuals that have been subjected to laboratory tests and selection consisted of three groups:

1. A group of individuals randomly selected from the generative progeny of a stand under standard forest management. These trees have been given the following symbols: PSI-6, PSI-9, PSI-10, PSI-13.

2. A group of individuals which are the vegetative progeny of plus trees originating from various parts of Poland, that is trees the selection differential of which relative to mother stands was not less than 3.48δ

$\left(i = \frac{x - \bar{x}}{\delta} > 3.48\delta\right)$ for all characters jointly (Wright, 1962). They have numbers with letters K.

3. Generative progeny obtained from open pollination of trees mentioned in group 2.

In our studies detached pine shoots were subjected to the action of SO₂ after being placed in vessels with water as well as whole seedlings from seeds collected in our seed orchard. From the selected trees a list of which is given in table 1 eight shoots were cut under water and placed into a chamber. So prepared material, consisting of 4 shoots from each tree, have been placed in the control chamber and in the chamber where

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a 1 or 2 ppm concentrations of SO_2 acted upon the plants. Each clone was represented by material collected from 2 or 3 grafts. Mean light intensity in the control and gas chambers was 9500 Lx — 10 500 Lx.

In order to compare the degree of resistance to the action of SO_2 , transmittable to half-sib progeny seeds were collected from trees of the studied clones. Seedlings raised from this seed in a greenhouse when about 10 months old and 8 - 12 cm high have been subjected to the action of SO_2 .

The degree of damage of the plants was estimated visually using a six point scale according to Schönbach and others (1968). In this scale 0-means no visible damages on the leaf surface, and 5-means visible injuries to over 70% of the leaf surface. The concentration of SO_2 in the chamber was estimated using an intermediate method through absorption of the gas in a solution of sodium mercuric chloride, and a colorimetric determination of the SO_3^{2-} ions (West, Gaekke, 1956), as well as by the use of an SO_2 analyser produced by Juncalor Dessau (GDR). The doses of the gas used were 2 ppm for shoots and 1 ppm SO_2 for seedlings.

Genotypes of the analysed trees were investigated through the variance of L-Leucine aminopeptidase (LAP-3.4.1.1). For this purpose we used tissue of the macrogametophyte and homogenized it in a phosphate buffer of pH 7.5 with the addition of 5 mM of sorbic acid, 5 mM of cystein and 5 mM of saccharose. The homogenate was separated by means of starch gel electrophoresis in a modified discontinuous buffer systems (Poulik, 1957). For the enzyme separation a direct electrical current at a potential of 15V/cm was used. To visualize the location of LAP isoenzymes contained after electrophoresis in different parts of the gel, we used a buffer of tris-maleate pH 5.4 and a 0.5 mol concentration. The typical substrate for this enzyme is L-leucine-2-naphtylamide-HCl, nad Black K Salt for the staining of bands. For the details of methods see Bergmann (1973), Mejnartowicz and Bergmann (1975), Mejnartowicz (1976).

RESULTS

On the obtained electrophograms the occurrence of isozymes was seen in two zones. Similar results with the same enzyme have been obtained in the studies on Norway spruce, black pine and European larch (Bergmann 1973, Mejnartowicz and Bergman, 1975; Mejnartowicz, 1976; Nikolić and Bergmann, 1974). The LAP-A zone has molecules of a greater electrophoretic mobility which gather near the front, while in the LAP-B zone there gather the molecules with a lower electrical load. On the basis of the studies conducted so far on populations

of Scots pine from northern and central Poland a zymogram was drawn which is presented in Fig 1.

Studies on independent trees have shown that between zones there are segregations at a ratios of 1 : 1 in heteromorphic individuals. Deviation from the 1 : 1 ratio judged by the chi-square test were found to be insigni-

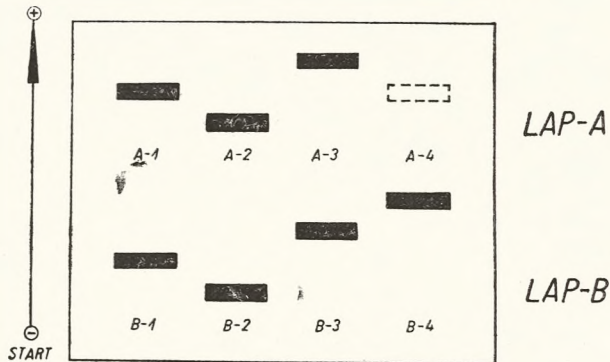


Fig. 1. LAP zymogram from *Pinus silvestris* macrogametophyte

ficant. In the studies we have found the occurrence of homozygous and heterozygous individuals (Table 1). Heterozygotes both in the LAP-A and LAP-B zones i.e. K-01-57, K-08-02 as well as many of the double homozygotes have had uninjured needles by SO_2 throughout the year. However among the studied pines in the group of heterozygotes and homozygotes individuals were also found having a variable degree of needle injury during a year.

The results of comparison of the differentiation of Scots pine individuals with respect to resistance to SO_2 and the differentiation of genotypes are given in Table 1.

We have for some pine individuals a full annual cycle of studies concerning their sensitivity to SO_2 . It was found that there is a considerable differentiation of the degree of needle injury by SO_2 to Scots pine trees in various parts of the season. During the winter period the needles were more resistant, even to high doses of SO_2 while in the summer and early autumn they were characterized by a greater sensitivity to this gas action (Table 1).

DISCUSSION

On the basis of presented data it was possible to study the injurious effect of sulphur dioxide on selected genotypes of Scots pine identified by the variability in L-leucine aminopeptidase loci. The population of double homozygotes is much more numerous than the heterozygous one,

which would tend to indicate that pine is characterized by a considerable homozygosity with respect to the LAP isozymes.

On the basis of the results of studies presented in table 1 it can be seen that there is no relation between the sensitivity to SO₂ action and the

Table 1

Characterisation of LAP genotypes of Scots pine with respect to resistance to SO₂ action

Isozyme i density	Period of SO ₂ treatment					seedlings 1977 18 - 21 IV
	shoots 1976					
	27 - 29 VII	11 - 13 VIII	18 - 20 VIII	24 - 26 VIII	3 - 6 XI	
Mean index injury						
Heterozygote at LAP-A locus						
K-01-57	0.7a			1.5a	0.0a	
PSI-6	2.6b	1.2a	1.5a	3.1b	0.4a	
K-01-82		1.6a	2.4b			0.8b
K-10-18	3.1b			3.6b	0.0a	
K-07-22		2.2b	2.9c			0.2a
Heterozygote at LAP-B locus						
K-08-02	0.9a			1.3a	0.1a	
K-01-02	1.6b			2.1a	0.6a	
K-07-01	1.9b			4.1b	0.1a	
Double heterozygote						
K-08-04 (A)	2.8b			3.1b	0.2a	
Double homozygote						
K-01-22	0.2a	0.6a	0.4a	0.9a	0.1a	0.7a
K-14-12		0.3a	0.3a	0.1a		0.1a
PSI-9	0.9a	0.8a	0.8a			
K-01-73		1.1a	0.5a			0.3a
PSI-13	0.3a	1.3a	0.6a	1.4a	1.0a	
K-14-13		1.6a	1.5b			0.2a
K-01-84		2.0b	2.2b			0.2a
K-10-03		2.1b	2.5c			0.5a
K-08-13		2.2b	2.3b			0.6a
K-01-16	2.1b	2.4b	3.1c	4.4b	2.9b	3.1c
K-07-04		2.6b	3.1c			0.3a
K-08-06		2.6b	3.6c			0.6a
K-14-10		2.7b	2.5c			2.5c
K-07-16		2.7b	2.8c			1.0b
K-14-14		2.8b	1.5b			1.5b
K-11-03		3.8b	2.8c			2.3c
K-08-05		3.9b	2.2b			1.6b
PSI-10		4.9b	3.0c			

The letters: a, b and c indicate that the results belong to a common undifferentiated group at 0.05 level of significance.

degree of homo- or heterozygosity of Scots pine LAP-genotypes. In these studies individuals with needles that are fairly resistant to the action of SO₂ were found to come both from the groups of heterozygotes and from the double homozygotes.

It was also found that there is a great variability in toxicity to the injurious gas depending on the season.

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LEON MEJNARTOWICZ, STEFAN BIAŁOBOK, PIOTR KAROLEWSKI

Genetyczne cechy osobników sosny zwyczajnej odpornych i wrażliwych na działanie SO₂

Streszczenie

Badano zmienność uszkodzeń powierzchni igieł sosny zwyczajnej powodowanych przez kontrolowane dawki SO₂. Materiałem do badań były gałązki, zebrane ze szczytów drzew doborowych oraz ze standardowego drzewostanu, a także potomstwo generatywne z niektórych szczepów drzew doborowych, powstałe z wolnego zapylania. Dla większości badanych drzew określiło ich genotypy na podstawie analizy zmienności enzymów leucyloaminopeptydazy (LAP) w tkance gametofitów żeńskich. W badanej grupie (około 20 sosen) przeważały osobniki homozygotyczne w obydwu wykrytych loci kodujących LAP.

Nie stwierdzono wyraźnej zależności pomiędzy stopniem heterozygotyczności drzew w odniesieniu do LAP-loci a stopniem odporności tych drzew na działanie SO₂.

Badając zmienność stopnia uszkodzeń igieł w ciągu roku stwierdzono największą odporność drzew w okresie późnojesiennym i zimowym, najmniejszą zaś wiosną i latem.

ЛЕОН МЕЙНАРТОВИЧ, СТЕФАН БЯЛОБОК, ПЕТР КАРОЛЕВСКИ

Генетические черты устойчивых и чувствительных к действию SO₂ особей сосны обыкновенной

Резюме

Исследовалась изменчивость повреждения поверхности хвои сосны обыкновенной, вызываемая действием контролируемых доз сернистого ангидрида. Материалом для исследования служили побеги, собранные с прививочной плантации плюсовых деревьев и со стандартного древостоя, а также генеративное потомство, возникшее в результате свободного опыления, с некоторых клонов плюсовых деревьев. Для большинства исследуемых клонов были определены их генотипы путем анализа изменчивости ферментов лейцинаминопептидазы (LAP) в тканях женских гаметофитов. В исследуемой группе ок. 20 сосен в большинстве представлены были гомозиготные особи в обеих обнаруженных локусах кодирующих LAP.

Не обнаружено четкой зависимости между степенью гетерозиготности деревьев в отношении LAP — локусов и степенью устойчивости этих деревьев к действию SO₂.

Исследуя изменчивость степени повреждения хвои в течение года, была обнаружена самая большая ее устойчивость поздней осенью и зимой, а самая незначительная весной и летом.