Investigation of Douglas-fir provenance test in North-Western Bulgaria at the age of 24 years

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ABSTRACT: A comparative analysis of growth rate and health condition of 54 Douglas-fir (*Pseudotsuga menziesii* [Mirb.] Franco) provenances was conducted at the age of 24 years. The provenance test was established in 1990 with 3-year-old (3+0) seedlings planted in a 2 × 2 m plot design and two replications. The provenances which were studied originated from North America and were separated into coastal, continental and Western Cascade groups. In 2011 the growth rate was studied in terms of height, diameter at breast height and stem volume. The health condition was assessed by the evidence of symptoms and degree of defoliation caused by the fungi *Phaeocryptopus gaeumannii* (Rohde) Petrak and *Rhabdocline pseudotsugae* (Syd.). The provenances Newhalem, Darrington, Idanha and Bremerton were characterized by the most rapid growth, highest productivity and lower susceptibility to both fungal pathogens. These Douglas-fir provenances were recommended for future afforestation in Bulgaria. Ten continental provenances should be excluded from future afforestation because of their lowest growth rate and productivity and high susceptibility to *P. gaeumannii* and *R. pseudotsugae*.

Keywords: adaptation; *Phaeocryptopus gaeumannii*; provenances; *Pseudotsuga menziesii*; *Rhabdocline pseudotsugae*; tree growth

Douglas-fir (*Pseudotsuga menziesii* [Mirb.] Franco) has been cultivated as a valuable coniferous tree species in Europe since 1827. The ecological and economic relevance of the species has been long paid attention all over the world due to its rapid growth and high productivity (Göhre 1958; Fenton 1967; Aussenac 1980; Binkley 1983; Oswald 1984; Hermann, Lavender 1999; Dunbar et al. 2002). In recent years the interest in Douglas-fir has been related to the established theories of climate changes and the possibilities of the species to adapt itself to predicted changes (St. Clair, Howe 2007; Martin 2011; Eilmann, Rigling 2012; Leites et al. 2012; Eilmann et al. 2013; Bastien et al. 2013). The main focus of species investigations was placed on the choice of an appropriate provenance for the specific country sites. As observed in many regions of Central and Southeast Europe during recent decades, the provenances from the western sites of the Cascade Mountains and from coastal seed zones in the states of Washington and Oregon have been recommended (Lavadinović et al. 2008; Weller 2010; Eilmann et al. 2013).

The introduction of Douglas-fir into Bulgaria began more than a century ago but it became widespread in the late 1950s and during the 1960s. The current area of the species plantations totals about 7,372 ha. The purposeful provenance studies on Douglas-fir initiated in 1987 are based on seed collections from 55 USA provenances. In 1990, 3-year-old seedlings of these provenances were established in plantations in three regions in Bulgaria (Popov 1995, 2001; Petkova 1999). The results from earlier studies emphasized that the coastal provenances and the provenances from the Western Cascade Mountains (Washington and Oregon) had the best growth rate (Popov 1995, 2001; Petkova 1999). At the age of 17 years, damage by needle cast diseases, caused by the ascomycete fungi *Phaeocryptopus gaeumannii* (Rohde) Petrak and *Rhabdocline pseudotsugae* (Syd.), was determined in established provenance tests

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(Georgieva 2009). Distinct differences between provenances comparing the degree of their susceptibility to both fungal pathogens were observed (Georgieva, Rossnev 2008; Georgieva 2009).

As it is important to consider sustainable plantation forest production, the objective of the present study was to analyse the growth rate and health condition of 54 Douglas-fir provenances at the age of 24 years in the test which is already established in the north-western part of Bulgaria.

MATERIAL AND METHODS

The Douglas-fir provenance test is located on the lower northern slopes of the Western Balkan Mountains (Northwest Bulgaria) in the Training Forest Enterprise in Petrohan. The trial was established in a flat terrain facing the east, at an altitude of 600 m, latitude of 43°14'N and longitude of 23°09'E. The soil is Orthic luvisol (FAO), mixed sandy and clay, slightly stony and very deep. The site is medium rich to rich. The climate in the region is temperate with an average annual temperature of 10.2°C and annual precipitation of 1,004 mm. The duration of the vegetation period is about 6 to 6.5 months.

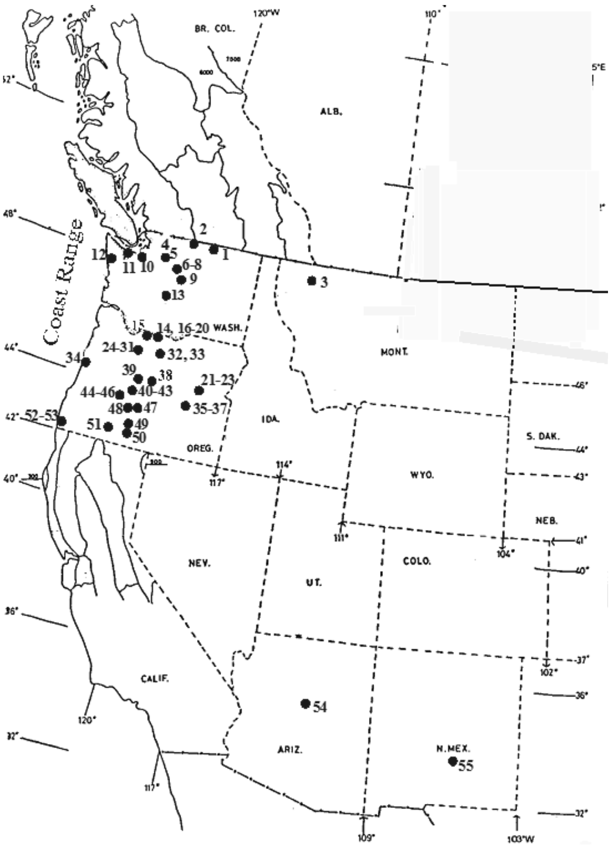

Fig. 1. Map of the geographic distribution of tested provenances in North America (Georgieva 2009)

The provenance test was established in the spring of 1990 with 3-year-old seedlings (3 + 0), which were planted in a 2 × 2 m plot design in two replications. Forty to forty-eight seedlings were planted on each plot. The 54 studied provenances originate from natural stands of Douglas-fir in North America (Table 1; Fig. 1) and were classified into three groups: continental (26), Western Cascade Mountain (22) and coastal (6) provenances.

In 2011, at the age of 24 years, the following indices were determined in all provenances: average diameter at breast height (DBH), average height and stem volume. Height and DBH were measured only in the first replication because a lot of individuals in the second replication had been lost due to unfavourable environmental conditions, namely excessive moisture and swamping. The average DBH for each provenance was determined after measuring the diameters of all trees on the plot in the following way. For each tree the mean diameter was obtained as the arithmetic mean of the measured two mutually perpendicular diameters to the nearest 0.1 cm. The obtained values were grouped in diameter classes by 2 cm. The average diameter for each provenance was calculated according to Eq. 1:

$$D = \sqrt{\frac{4G}{\pi N}} \text{ (cm)} \qquad (1)$$

where:

G – total basal area of the trees in the provenance ($G = \Sigma n_i \times g_i$ [m^2]),

n_i – number of trees in the respective diameter class (by 2 cm),

g_i – basal area in the respective diameter class (m^2),

N – number of trees in the provenance.

The average height (H) of each provenance was calculated as the arithmetic mean of the heights of 15–25 individual trees, distributed into diameter classes (by 2 cm).

The stem volume was calculated according to Eq. 2:

$$V = G \times HF \text{ (m}^3\text{)} \qquad (2)$$

where:

G – total basal area of trees in the provenance (m^2),

HF – height form factor for *Abies alba* Mill. (Poryazov et al. 2004).

The statistical program SPSS 11 (IBM, New York, USA) was used for analysing the variable heights and DBH's. Statistical parameters (arithmetic mean, standard deviation and standard error) were assessed for the analyed height and DBH per provenance. The differences between provenances were tested by analysis of variance (ANOVA). In cases

Table 1. Geographical distribution of Douglas-fir provenances

Provenance group	Provenance number	Seed zone	Provenance name	Geographical coordinates (N)	(W)	Altitude (m)	Number of trees
Continental							
Montana	3.	Montana	Whitefish	48.5	114.5	1,050	36
New Mexico	55.	840	Alamogordo	33.0	105.8	750	14
Washington	1.	612	Greenwood	49.0	119.0	1,350	37
	2.	600	Keremeos	49.0	120.0	750	33
	13.	641	Naches	46.5	121.3	1,050	28
East Cascade Mountains							
Oregon	14.	661	Parkdale	45.5	121.5	1,650	26
	15.	661	Parkdale	45.5	121.7	1,500	29
	16.	661	Parkdale	45.5	121.5	1,350	26
	17.	661	Parkdale	45.5	121.5	1,200	28
	18.	661	Parkdale	45.5	121.5	1,050	30
	19.	661	Parkdale	45.5	121.5	900	30
	20.	661	Parkdale	45.5	121.5	750	28
	33.	662	Warm Springs	45.0	121.5	667	34
	32.	662	Warm Springs	45.0	122.0	900	28
	38.	675	Santiam Pass	44.3	121.6	1,125	32
	47.	681	Crescent	43.3	121.8	1,650	28
	48.	681	Crescent	43.3	122.0	1,500	30
East Oregon	21.	863	Bates	45.0	118.5	1,667	18
	22.	863	Bates	45.0	118.5	1,500	30
	23.	863	Bates	45.0	118.5	1,333	23
	35.	892	Canyon City	44.5	119.0	1,500	29
	36.	892	Canyon City	44.5	119.0	1,350	27
	37.	892	Canyon City	44.5	119.0	1,650	14
Coastal Mountains	49.	501	Crater Lake	42.7	122.5	1,200	29
South Oregon	50.	501	Medford	42.5	122.5	1,050	25
	51.	502	Medford	42.6	122.8	900	24
Western Cascade Mountains							
Washington	4.	402	Newhalem	48.5	121,5	667	22
	5.	402	Newhalem	48.5	121.5	500	29
	6.	403	Darrington	48.0	121.5	1,167	27
	7.	403	Darrington	48.0	121.5	1,000	26
	8.	403	Darrington	48.0	121.5	833	23
	9.	411	Monroe	47.8	121.3	525	30
Oregon	24.	452	Idanha	45.0	122.0	1,050	29
	25.	452	Idanha	45.0	122.0	1,200	33
	26.	452	Idanha	45.0	122.0	1,050	31
	27.	452	Idanha	45.0	122.0	900	29
	28.	452	Idanha	45.0	122.0	1,333	27
	29.	452	Idanha	45.0	122.0	750	32
	30.	452	Idanha	45.0	122.0	750	32
	31.	452	Idanha	45.0	122.0	750	29
	40.	472	Oakridge	44.0	122.0	1,667	24
	41.	472	Oakridge	44.0	122.0	1,500	26
	46.	472	Oakridge	43.8	122.5	1,500	29
	39.	473	Santiam Pass	44.3	121.8	1,500	28
	42.	473	Oakridge	44.0	122.0	1,333	28
	43.	482	Oakridge	44.0	122.0	900	30
	44.	482	Oakridge	43.8	122.5	1,350	31
	45.	482	Oakridge	43.8	122.5	1,200	25
Coastal							
Washington	12.	12	Moclips	47.5	124.0	600	27
	10.	222	Bremerton	47.7	123.0	600	30
	11.	222	Bremerton	47.7	123.5	450	29
Oregon	34.	53	Toledo	44.6	123.8	150	32
	52.	82	Brookings	42.0	124.5	833	20
	53.	82	Brookings	42.0	124.5	667	20

Table 2. Defoliation classes

Damage class	Needle loss (%)	Degree of defoliation
0	0–10	no needle defoliation
1	> 10–25	slight defoliation
2	> 25–60	moderate defoliation
3	> 60–100	severe defoliation
4	100	completely defoliated

where there was a significant statistical difference between provenances, the provenances which differed between themselves were determined by using the post hoc LSD test.

In the provenance test, the health condition of 25 randomly selected trees in each provenance was surveyed just prior to bud break, to evaluate the occurrence of symptoms of needle cast diseases. The occurrence of needle cast disease was revealed by the presence of reproductive fungal structures, defoliation and discoloration of needles in the crowns of infected trees. The extent of visually assessed defoliation of trees, as developed by the International Co-operative Programme (ICP Forests) of the Executive Committee for the Convention on Long-range Transboundary Air Pollution in Europe was used (Eichhorn et al. 2010). Five defoliation classes were defined (Table 2). Defoliation of more than 25% was taken as a threshold for categorizing a tree as damaged.

The average defoliation of each provenance was expressed as the arithmetic mean of the values of defoliation of all 25 trees in percentage per provenance level. According to scored average defoliation each provenance was defined as: slightly susceptible (with no or slight defoliation); moderately susceptible (with moderate defoliation) and heavily susceptible (with severe defoliation or completely defoliated) (Chumakov 1974).

RESULTS AND DISCUSSION

The overall mean height at the age of 24 years was 14.0 m. The highest was provenance 6 (Darrington) of the Western Cascade Mountains with average height 20.0 m. Provenances 4 and 5 (Newhalem), 7 (Darrington), 9 (Monroe), 27, 28 and 29 (Idanha) from the Western Cascade Mountains and coastal provenance 11 (Bremerton) were defined as rapidly growing too with an average height of 17–18 m (Fig. 2a). The value for the height growth of these provenances exceeds the value for the first site index (15.5 m at the age of 25 years) according to tables of Bergel (1985). These provenances were characterized as most rapidly growing in height in the same provenance test at 20 years of age (Petkova 2011). The provenances Newhalem, Darrington and Idanha were defined as the most rapidly growing in height for the test in the south-western part of Bulgaria (Popov 2011). The provenance Darrington was described as stable in a wide range of site conditions and was recommended as a seed source for the conditions of Austria (Bastien et al. 2013). Ten continental provenances 1 (Greenwood) and 2 (Keremeos) of Washington, 3 (Whitefish) of Montana, 21, 22, 23 (Bates), 35, 36, 37 (Canyon City) of Eastern Oregon and 55 Alamogordo of New Mexico were registered with the slowest growth in height (Fig. 2a).

The average DBH of the provenance test at the age of 24 years was 16.0 cm. The largest average diameter was measured in provenance 4 (Newhalem) of the Western Cascades – 20.2 cm (Fig.2b). Provenances 15, 16 and 17 (Parkdale), 38 (Santiam Pass), 47 (Crescent) of East Cascade, 50 and 51 (Medford) South Oregon, 5 (Newhalem), 6 and 7 (Darrington), 30 (Idanha) of Western Cascade and coastal provenances 52 and 53 (Brookings) were also characterized by very good growth in diameter (average diameter of about 18 cm and over). Continental provenances 1 (Greenwood) and 2 (Keremeos) of Washington, 3 (Whitefish) of Montana, 21, 22, 23 (Bates), 35, 36, 37 (Canyon City) of Eastern Oregon and 55 Alomogordo of New Mexico were recorded with very low growth in diameter.

By applying the post hoc LSD test a high statistical significance by factor provenance ($P < 0.001$) regarding both height and diameter was established (Table 3).

For the entire provenance test, the average value of stem volume was 237 $m^3 \cdot ha^{-1}$. The highest stem volume was calculated for provenances 5 (Newhalem) – 385 $m^3 \cdot ha^{-1}$ and 6 (Darrington) – 376 $m^3 \cdot ha^{-1}$ of the Western Cascade. Provenances 15, 17 and 18 (Parkdale) and 47 (Crescent) from East Cascade,

Table 3. Results of ANOVA analyses

Source of variation	df	Mean squares	*F*	*P*
ANOVA for the height				
Provenance	51	79.256	35.753	0.000
ANOVA for the diameter				
Provenance	51	243.471	14.016	0.000

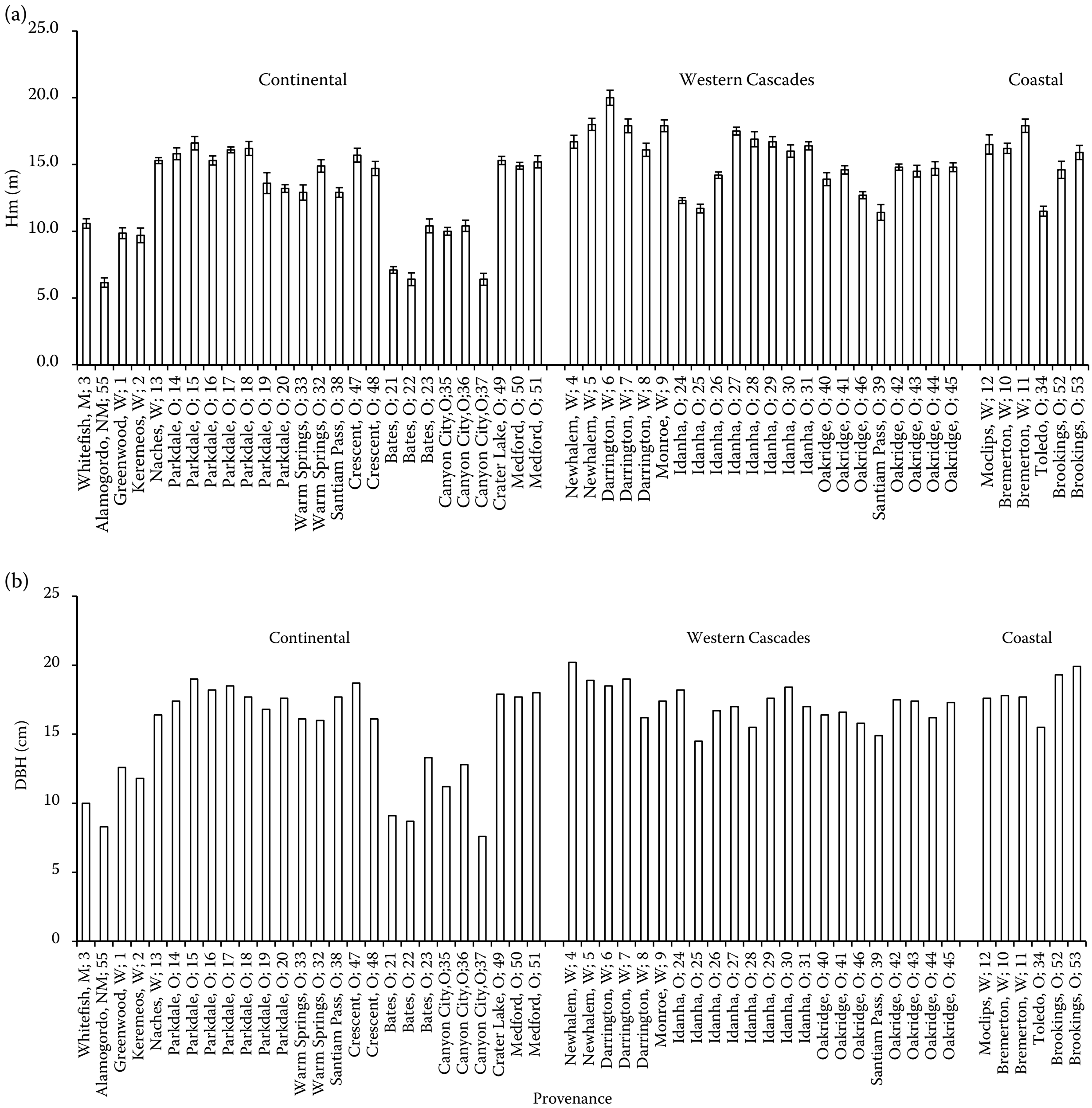

Fig. 2. Average height (a), diameter at breast height (b) of 24-year-old Douglas-fir provenances planted in 1990 in NW Bulgaria

4 (Newhalem), 7 and 9 (Darrington), 27, 29 and 30 (Idanha) and 43 (Oakridge) from Western Cascade and the coastal 10 and 11 (Bremerton) deserve particular attention. Their stem volume exceeded 300 $m^3 \cdot ha^{-1}$, which confirms the great potential and perspective of these Douglas-fir provenances.

According to Popov (2013) provenances 4 (Newhalem), 7 (Darrington) and 29 (Idanha) were the most productive in the other provenance test established in south-west Bulgaria.

Studies of Weller (2012) for Douglas-fir provenance test at the age of 38 years in northwestern Germany demonstrated that provenances Darrington differed in improved establishment and outstanding productivity. The slowest growing continental provenances in height and diameter had the lowest productivity.

In 2011 symptoms and damage of Swiss needle cast caused by the ascomycete fungi *P. gaeumannii* were detected on all Douglas-fir trees across all provenances. Damage caused by *R. pseudotsugae* was recorded only on trees which originated from continental areas. The first observation of these pathogens in the same provenance test was made at the age of 17 years in 2004 (Fig. 3a). The 44 provenances from the coastal area and from the Cascade Mountains were characterized by no or slight defoliation and ten provenances from the continental area were assessed as highly susceptible both to *P. gaeumannii* and to *R. pseudotsugae* (Georgieva 2009). The infection rates significantly

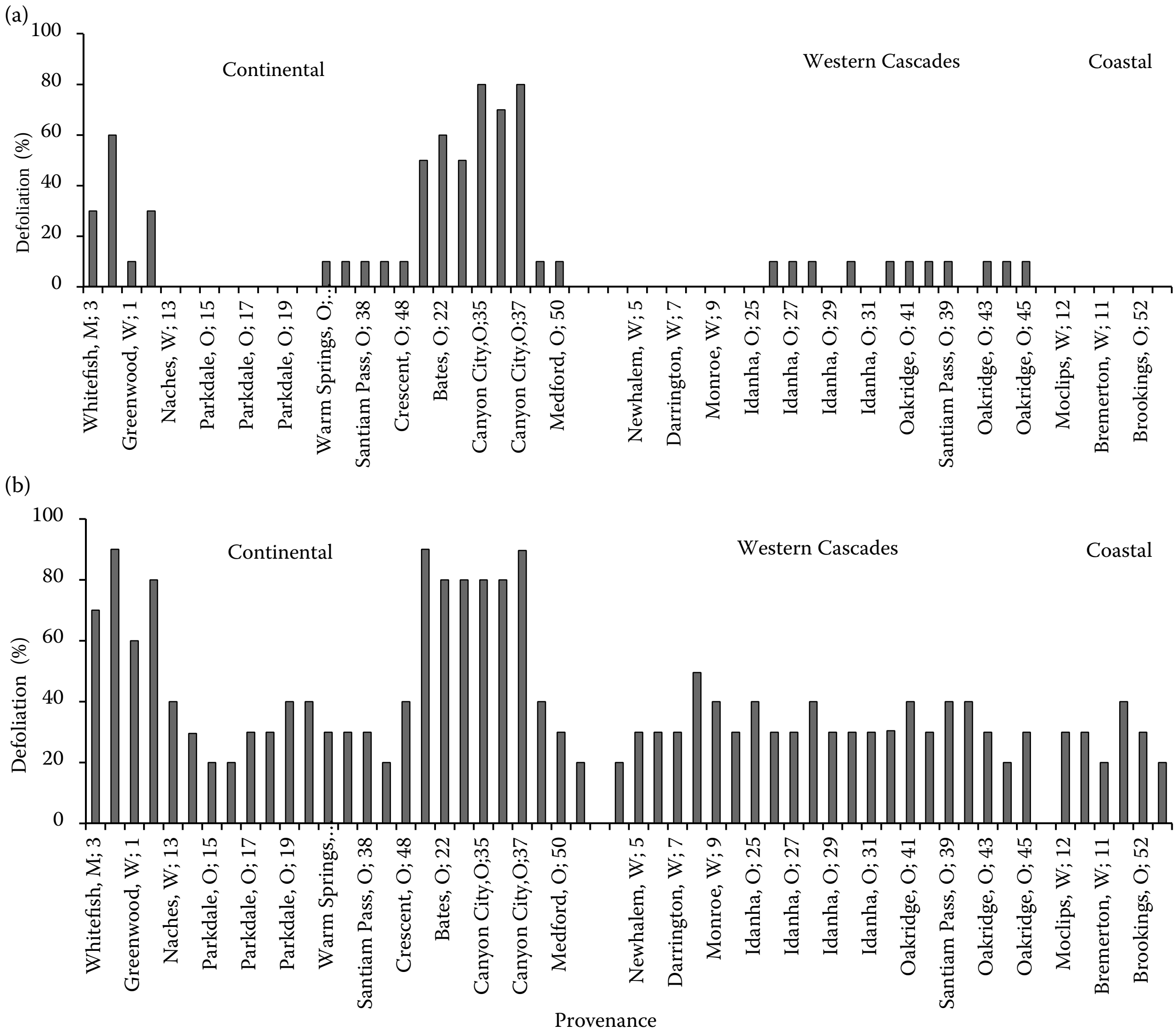

Fig. 3. Distribution of average present defoliation defined in 2004 (a), 2011 (b) in 54 provenances

increased with years, and in 2011, the proportion of Douglas-fir trees infected by *P. gaeumannii* reached 100% in all 54 provenances (Fig. 3b). All assessed trees were defined as susceptible to the pathogen because of the presence of numerous fruit structures (pseudothecia) detected on needles resulting in yellowing discoloration and an increasing percentage with crown defoliation. During the recent decades, outbreaks of Swiss needle cast have occurred in both native and plantation grown *P. menziesii* in the Pacific Northwest (Hansen et al. 2000; Manter et al. 2005) and forest plantations in Europe, Turkey (Temel et al. 2003), Australia (Marks et al. 1989) and New Zealand (Hood et al. 1990; Watt et al. 2010). According to Hood et al. (1990) no provenance is immune to the disease but the provenances from the coastal zones are slower to become infected than provenances from continental sites.

A high variability was observed between provenances from the continental area concerning their height growth (Fig. 2a) and susceptibility to needle cast diseases (Fig. 3b). The seriousness of the needle cast disease complex was emphasized in the ten provenances from continental areas: 1 (Greenwood) and 2 (Keremeos), 3 (Whitefish), 21, 22, and 23 (Bates), 35, 36, and 37 (Canyon City) and 55 Alomogordo. These were characterized by symptoms of increasing defoliation, decreasing vitality after infection, the slowest growth and the lowest productivity (Figs 2 and 3b).

It was observed that the density of pseudothecia increased with needle age, and increasing proportions of stomata occupied by pseudothecia were associated with increasing amounts of defoliation. Severely defoliated trees from the ten continental provenances retained only 1-year-old needles. The worst health condition of these trees was a consequence of heavy susceptibility to both fungal pathogens – *P. gaeumannii* and *R. pseudotsugae*. A major effect of the disease was a reduction of needle area in moderately susceptible trees and the death of heavily susceptible trees. Similar results of slow growth and heavy susceptibility to needle cast disease were obtained on trees from the same ten provenances as in the other established provenance tests in Bulgaria (Popov 2001; Georgieva 2009; Popov 2012).

The trees of the remaining 16 continental provenances were assessed as being with slight to moderate defoliation, the average percentage of defoliation varying between 20% and 40% (Fig. 3b). The 4-year-old needles on these trees were retained on branches and had very low levels of pseudothecia on the 1-year-old needles.

The provenances from the Western Cascades Mountains and from the coastal area were defined with slight (20%) to moderate (50%) defoliation but no significant differences in the average percent of defoliation were ascertained among them. In 2011 an increase in the degree of defoliation was determined in these provenances as compared to the assessment made in 2004 (Fig. 3a). In 2004 all coastal provenances and 17 provenances from the West Cascade Mountains were not defoliated by *P. gaeumannii.* The trees from the remaining West Cascade Mountain provenances were up to 10% defoliated (Fig. 3a).

The distribution of damage classes over all trees is shown in Table 4. Nearly one third (31%) of the trees from continental provenances were dead and 7.7 % were characterized by a degree of defoliation higher than 60%.

The provenances from the Western Cascade Mountains and the coastal area were defined as slightly to moderately susceptible. However, the proportion of damaged trees (degree of defoliation higher than 25%) was significant – 90.9% in Western Cascade Mountains and 66.7% in coastal provenances (Table 4). According to Hood et al. (1990), Kleinschmit and Svolba (1997), Liesebach (2010), Bastien et al. (2013) the provenances from the Western Cascades Mountains and coastal area were slower to become infected than provenances from continental sites.

Table 4. Frequency of Swiss needle cast damage classes for three provenance groups

Provenance area	Total no. of trees	Damage classes				
		0	1	2	3	4
Continental	650	–	15.4	46.1	7.7	30.8
Western Cascade Mts.	550	–	2.1	90.9	–	–
Coastal	150	–	33.3	66.7	–	–

In 2004 the Douglas-fir trees from all 54 provenances were examined during the period of bud burst and elongation of new needles and shoots (Georgieva 2009). It was found that there are differences between the provenances in terms of their bud-burst phenology. The provenances from the continental area began to burst during the last week of April. With the other origins (from the Cascade Mountains and the coastal areas) bud burst and elongation of new needles began in the middle of May. According to Boyce (1961) the lower susceptibility of the provenances from the Cascade Mountains and coastal areas to needle cast diseases was due to the later development of buds and the formation of new shoots – after the massive dissemination of ascospores of the fungus.

The evidence for growth stress as a primary cause of the decline showed that in many unthinned stands this led to conditions which certainly allowed the fungal pathogen *P. gaeumannii* to have an increased effect. In order to limit the further dissemination of the disease in 2008 the provenance test was thinned and dead and high affected trees were removed. The purpose of adopting this type of regime was to improve airflow thus preventing the conditions on which the fungus thrives and so making the trees less susceptible to fungal diseases.

CONCLUSIONS

The provenances Newhalem, Darrington, Idanha from the western Cascade Mountains and coastal provenance Bremerton were characterized by the most rapid growth and highest productivity in Douglas-fir provenance test in NW Bulgaria at the age of 24 years. They were less susceptible to the Swiss needle cast disease caused by *P. gaeumannii* and *Rhabdocline* needle cast caused by *R. pseudotsugae*. From our perspective these provenances can be recommended for the future afforestation with Douglas-fir in Bulgaria due to expectation of their rapid growth, high productivity and lower susceptibility to fungal diseases.

The slowest growth and the lowest productivity were ascertained in the continental provenances Greenwood and Keremeos of Washington, Whitefish of Montana, Bates and Canyon City of eastern Oregon and Alamogordo of New Mexico, which were defined as the highest susceptible to *P. gaeumannii* and *R. pseudotsugae.* These provenances should be excluded from future afforestation with Douglas-fir in Bulgaria.

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