Analysis of herbicide effects on Douglas fir (*Pseudotsuga menziesii* [Mirb.] Franco) natural regeneration

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ABSTRACT: Douglas fir (*Pseudotsuga menziesii* [Mirb.] Franco) natural regeneration under parent stand after weed suppression was investigated in the area of the Training Forest Enterprise in Kostelec nad Černými lesy. The study evaluates two measurements made in a two-year interval. The parent stand, where Douglas fir grows in mixture with our domestic tree species, is one of the oldest and most productive stands at the Training Forest Enterprise in Kostelec nad Černými lesy. Herbicides for the control of weeds were applied onto three permanent experimental plots under the parent stand. Measurements done on the plots show a high potential of natural regeneration in areas where the negative impact of forest weeds was suppressed. This conclusion is acknowledged by the high seedling number found on research plots treated with herbicides. The number of Douglas fir seedlings varied from 21,600 per hectare counted on the plot treated with Dominator to 26,650 seedlings on the plot with Velpar treatment even six years after the soil preparation. Statistical analysis confirmed that the seedling number on the plot without any chemical preparation was significantly lower, only 950 individuals per hectare (χ^2 = 926.84, df = 3, P << 0.01).

Keywords: Douglas fir; herbicides; introduced tree species; natural regeneration

Importance and possibilities of tree species introduction

Introduced tree species could take an important place in sustainable multifunction forest management in the Czech Republic, similarly like in other European countries. The introduction has a long tradition in the Czech lands. Tree species have been introduced not only for aesthetic purposes but also for their wood and non-wood-producing functions. Further, thanks to their high potential, they are hardly replaceable (Hart, Remeš 2006). The most actual problem is to determine the position of the introduced tree species in the present forest management, it means to confirm or to overcome the assumption of their importance and utilization.

In the first half of the 20th century, Douglas fir growing was perceived as a possibility of increasing the forest productivity. On the contrary, at the end of the century, Douglas fir (*Pseudotsuga menziesii*) was seen very critically and near-natural forestry was preferred (Martiník, Kantor 2004). The actual experiences confirm that Douglas fir is one of the most productive tree species. This statement was mentioned by authors from the Czech Republic (Hofman 1964; Wolf 1998a,b; Kantor et al. 2001; Remeš 2002; Kantor, Kotlan 2006; Kantor 2008) as well as from the neighbouring countries (Huss 1996; Burgbacher, Greve 1996; Greguš 1996; Ponette et al. 2001). Mainly data on high growing stock, wood production and especially the values of mensurational indices attract attention. At present, Douglas fir grows in our forests on the area of 4,808.5 ha, that is about 0.2% of forest land of the Czech Republic. Forest stands of the first four age classes prevail (Anonymus 2008). Further, a favourable impact of this species on forest soil is expected (Podrázský 1998; Podrázský, Remeš 2006, 2008; Podrázský et al. 2009).

Introduction in the area of the Training Forest Enterprise in Kostelec nad Černými lesy has a long tradition and is mainly connected with Liechtenstein estate. On the area of 6,734 ha, Douglas fir occurs in 98 stands and its proportion ranges from 5 to 100%. Douglas fir reduced area is 14.56 ha, i.e. 0.22% of the overall stand area at the Training Forest Enterprise.

The provenance suitability for the import of high-quality seed has been discussed very often. According to Červenský (2001) the superior sources of the necessary amount of seed of an acceptable and well-tried provenance are sufficient. Hence, mainly domestic seed sources should be used and we should also take advantage of natural regeneration. According to a study worked up by the Forestry and Game Management Research Institute in Jíloviště-Strnady, Douglas fir could be regenerated and planted on up to 2% of the yearly regenerated area, i.e. on 400 to 500 ha (Šindelář 2003).

Natural regeneration

Natural regeneration is a native process taking place in forest development. It is the ability and product of the forest ecosystem self-reproduction. Nevertheless, we can consider natural regeneration to be a result of the purposeful and systematic work of a forest manager (Korpeľ et al. 1991). Bušina (2007a) stated that natural regeneration of forest tree species is known to be one of the fundamental and regular processes of the life cycle of each virgin forest and native forest. Natural regeneration is particularly important for the preservation of genetic resources of forest tree species populations. Further, the risk of the temporal or permanent deterioration of forest site conditions is much lower in comparison with artificial regeneration. Natural regeneration enables to grow high-quality stands because the number of naturally regenerated individuals is much higher than that of artificially planted ones and thus we have much more possibilities to choose superior individuals during stand tending. Natural regeneration is a notable part of near-natural forest management (Kupka 2002). It also contributes to a higher stability of forest ecosystems (Poleno 1997; Podrázský 1998).

Biological conditions for Douglas fir natural regeneration are favourable at most sites. The interval between seed years is 5–7 years, the beginning of fertility is at the age of 20 to 30 and it lasts till high age. Seeds germinate well in mineral soil. Most seeds fall to a distance of 300 m from the stand of generative Douglas fir trees (Úradníček, Chmelař 1995). Hofman (1964) stated that Douglas fir natural regeneration under the shelter of parent stand takes place quite frequently in most European countries. It appears mainly in properly tended stands at the age of 60 years. Problems with pollination may arise if Douglas fir occurs in small groups or is only interspersed. Further, a huge amount of dead seeds is present (Kinský, Šika 1987). Very little is known about parthenocarpy.

Šindelář (2003) and Šindelář and Beran (2004) reported that it is possible and convenient to regenerate high-quality Douglas fir stands in a natural way at favourable sites in conditions of the Czech Republic. For the success of natural regeneration they recommended to prepare hospitable conditions by formation of the crown canopy and by soil preparation during the seed year. According to Korpeľ et al. (1991) light is the most important factor after sprouting of seedlings. Bušina (2007b) concluded that the lateral light is much more important for the natural regeneration density than the light coming through the parent stand crowns. The study by Kinský and Šika (1987) confirmed that Douglas fir regenerated very well (abundance 400–5,000 individuals·ha^{-1}) mainly thanks to the lateral self-seeding, in conditions of the 3K forest type group. Coherent regeneration appeared even in places from which the nearest Douglas fir trees were 25 m distant. Wolf (1998a,b), who conducted his research work in the surroundings of the town of Písek, found out that Douglas fir natural regeneration is possible on acid soil with less aggressive weeds. Conversely, most seedlings died mainly during the second and third year as a consequence of weed competition on more nutrient-rich soils. Hart and Remeš (2006) presented their very good experiences with natural regeneration under parent stand at the Training Forest Enterprise in Kostelec nad Černými lesy after the application of herbicides that suppressed the negative influence of weeds on seedlings. The present study is the continuation and enlargement of such research. The aim of the study is to confirm the utility and relevance of chemical soil preparation in forest management.

MATERIAL AND METHODS

Study area

The area of the Training Forest Enterprise in Kostelec nad Černými lesy is located 25–50 km northeast of Prague. The altitude ranges from 210–538 m a.s.l. Climatic characteristics include the mean annual temperature 7.0–7.5°C and 13.0–13.8°C during the growing season. Growing season lasts 153 days on average. The mean annual precipitation amounts to 600–650 mm. Rainfall distribution during the year is favourable (65% of rainfall during the growing season). The wind direction is modified by the terrain to a great extent. West winds prevail, destructive winds from the southeast blow scarcely (Forest Management Plan 2001).

Forest stand 441D10 is situated in the Jevany forest district, 4 km southeast of Kostelec nad Černými lesy. It is located on a moderate northeastern slope. The main forest type is 4O1, management set of stands is 461. The stand size is 3.29 ha. The tree species composition is as follows: Norway spruce (*Picea abies* [L.] Karst.) 75%, Douglas fir (*Pseudotsuga menziesii* [Mirb.] Franco) 25%, other tree species silver fir (*Abies alba* Mill.), Scots pine (*Pinus sylvestris* [L.]), European larch (*Larix decidua* Mill.), sessile oak (*Quercus petraea* [Matt.] Liebl.) and European birch (*Betula verrucosa* Ehrh.) are interspersed. Douglas fir and Norway spruce fructify every year. The altitude of the stand is 410 m a.s.l. Stocking was 10 and the canopy was open at the time of the trial establishment. The age of the stand is 100 years, the canopy is open and stocking was 11 in 2008.

Three permanent research plots (PRP) were established under the parent stand. PRP are 0.04 ha square-shaped areas located 30 m from the forest edge. Each PRP is divided into 400 squares with 1 m long sides. The canopy is 79% on PRP 1.73% on PRP 2 and 64% on PRP 3. The seedling number was monitored in each square in 2006 and 2008. Survival of seedlings was figured out thanks to repeated seedling numeration and consequent determination of age. Seedling height and diameter at root collar were also monitored. The evaluation of these parameters will be the aim of another study.

Weed control

Three types of herbicides were applied onto PRPs (Table 1 shows the types, concentrations and amounts of the chemicals). Herbicides were used to minimize the weed competition and to enable natural regeneration. Before the soil preparation was done, several Norway spruce trees were felled to open the canopy and to increase the amount of incident light necessary for seedling growth. The herbicide application was realized in 2002. There were no weeds on all PRPs after the application of herbicides and the site conditions were also the same and thus we can consider that the initial conditions for seedling establishment were identical on all PRPs. The effects of herbicides on seedling growth could then be easily observed.

The investigation began four years after the application of herbicides to avoid the effects of residues of the herbicide active substances on seedlings. Two measurements carried out in a two-year interval (2006 and 2008) always after the end of growing season are mentioned in the present study. In 2008, one check measurement was done on a plot without soil preparation to find out the impact of herbicide application on natural regeneration. The number and species of seedlings were determined and also their age and height (their evaluation will be the subject of a further study).

Statistical analysis

The differences between PRPs were examined using the χ^2 test according to Zvára (2006):

$$\chi^2 = \sum_{i=1}^{r} \sum_{j=1}^{c} \frac{(N_{ij} - n_i \times N_j/n)^2}{n_i \times N_j/n}$$

where:
N_{ij} – observed seedling frequencies,
$n_i \times N_j/n$ – expected seedling frequencies,
r (c) – number of research plots (tree species).

The analyses were done for each year separately. If the calculated value exceeded the critical χ^2 value, the differences were considered as statistically significant (the degrees of freedom df used in the statistical evaluation are either 2 or 3 for a comparison of Douglas fir numbers between the particular plots,

Table 1. Chemical preparatives and concentration

Permanent research plot	Application date	Herbicide	Herbicide amount
1	2. 5. 2002	Velpar 90	3 kg/400 l water
2	2. 5. 2002	Roundup Forte	3.5 kg/200 l water
3	2. 5. 2002	Dominator	7 l /200 l water

Table 2. Seedling number on three PRPs after growing season 2006

Tree species	Seedling age					Seedling number per plot	Seedling number per ha
	1	2	3	4	5		
PRP 1							
DF	504	641	148	12	1	1,306	32,650
NS	2,650	85	30	3	10	2,778	69,450
SF	23	22	16	2	0	63	1,575
SP	10	4	2	0	0	16	400
Others	8	2	3	0	0	13	325
Σ per plot	3,195	754	199	17	11	4,176	
Σ per ha	79,875	18,850	4,975	425	275		104,400
PRP 2							
DF	287	306	119	11	2	725	18,125
NS	3,267	59	23	10	3	3,362	84,050
SF	22	30	11	1	1	65	1,625
SP	2	2	1	0	0	5	125
Others	7	1	2	0	0	10	250
Σ per plot	3,585	398	156	22	6	4,167	
Σ per ha	89,625	9,950	3,900	550	150		104,175
PRP 3							
DF	234	446	175	18	4	877	21,925
NS	993	43	8	4	1	1,049	26,225
SF	21	13	2	1	1	38	950
SP	6	1	2	0	0	9	225
Others	8	1	0	0	0	9	225
Σ per plot	1,262	504	187	23	6	1,982	
Σ per ha	31,550	12,600	4,675	575	150		49,550

PRP – permanent research plot, DF – Douglas fir, NS – Norway spruce, SF – silver fir, SP – Scots pine

the critical value is 5.9918 or 7.8153 for a 0.05 significance level; or 4 when comparing two PRPs, the critical value is 9.4884 for a 0.05 significance level; or 8 when comparing three PRPs, the critical value is 15.509 for a 0.05 significance level; or the df are 12 when comparing three PRPs and the check plot, the critical value is 21.028 for a 0.05 significance level).

RESULTS AND DISCUSSION

The chemical soil preparation that was applied onto PRPs and was to help the natural regeneration survival proved different effects. Nevertheless, this type of soil preparation seems to be very beneficial as was confirmed on the basis of previously published studies, e.g. by Harper et al. (2004), who observed results after the application of Velpar and another herbicide. We can observe the best results on PRP 1 and 2. Velpar, a selective herbicide, was used on PRP 1. Weeds occurred only sporadically and could not inhibit the growth of natural regeneration. 104,400 seedlings per ha (Table 2) were counted on PRP 1. Douglas fir accounted for 31.3%, nevertheless, Norway spruce seedlings were the most abundant (66.5%) and the other tree species accounted for 2.2%.

Another type of selective herbicide, Roundup Forte, was tested on PRP 2. Forest weeds appeared more frequently on this plot in comparison with PRP 1 and partially retarded the seedling development. Nevertheless, the overall weed incidence was scarce and could not essentially influence the growth of natural regeneration. The total seedling number, counting 104,175 per ha, was similar like on PRP 1 (Table 2). The proportion of Douglas fir seedlings was 17.4%. The proportion of Norway spruce seedlings 80.7% was higher than on PRP 1, and the proportion of other tree species was 1.9%.

The third PRP was treated with Dominator. The influence of weeds was obvious. The weed cover was so strong in more backlit places that it totally inhibited natural regeneration. Together, 49,550 seedlings per ha were found on PRP 3 (Table 2). The proportion of Douglas fir (44.2%) and Norway spruce (52.9%) seedlings was almost equal. Other species accounted for 2.9% seedlings.

Statistical analyses proved differences in the tree species frequency distribution on the particular PRPs in 2006 (χ^2 = 536.88, df = 8, $P << 0.01$). Furthermore, the incidence of Douglas fir seedlings is markedly distant on the particular plots (χ^2 = 187.31, df = 2, $P << 0.01$). The total number of Douglas fir seedlings is the highest on PRP 1 treated with Velpar (1,306; i.e. 32,650 seedlings per ha), lower on PRP 3 treated with Dominator (877; i.e. 21,925 seedlings per ha), and the lowest is on PRP 2 where Roundup Forte was applied (725; i.e. 8,125 seedlings per ha).

PRPs were repeatedly measured in 2008. The best results were obtained on PRP 1. Weeds occurred only seldom after Velpar application. Even though Harper et al. (2004) stated that the herbicide effec-

Table 3. Seedling number on three PRPs after growing season 2008

Tree species	Seedling age					Seedling number per plot	Seedling number per hectare
	1	2	3	4	5		
PRP 1							
DF	234	336	333	137	26	1,066	26,650
NS	346	1,170	360	35	30	1,941	48,525
SF	53	393	18	7	4	475	11,875
SP	7	1	0	0	0	8	200
Others	2	6	6	2	3	19	475
Σ per plot	642	1,906	717	181	63	3,509	
Σ per ha	16,050	47,650	17,925	4,525	1,575		87,725
PRP 2							
DF	565	350	87	15	2	1,019	25,475
NS	648	495	35	3	10	1,191	29,775
SF	311	146	15	3	0	475	11,875
SP	8	0	0	0	0	8	200
Others	2	2	1	0	0	5	125
Σ per plot	1,534	993	138	21	12	2,698	
Σ per ha	38,350	24,825	3,450	525	300		67,450
PRP 3							
DF	236	218	223	140	47	864	21,600
NS	242	289	69	13	4	617	15,425
SF	68	19	10	2	0	99	2,475
SP	1	1	0	0	1	3	75
Others	0	4	0	0	0	4	100
Σ per plot	547	531	302	155	52	1,587	
Σ per ha	13,675	13,275	7,550	3,875	1,300		39,675

PRP – permanent research plot, DF – Douglas fir, NS – Norway spruce, SF – silver fir, SP – Scots pine tive control of grass and herbaceous vegetation lasts for three to four years after Velpar application, this study confirmed that the negative influence of weeds on natural regeneration was weak even six years after Velpar application. The seedling number on PRP 1 was 87,725 per ha (Table 3). The proportion of Norway spruce was 55.3%, the proportion of Douglas fir 30.4% and that of other tree species 14.3%. The high number of surviving seedlings on herbicide-treated plots shows that conditions during the establishment of natural regeneration strongly affect the further seedling growth, as stated also by Newton and Preest (1988), who made experiments with Douglas fir seedlings. They found out that seedlings on plots with no herbaceous vegetation experienced less water stress and their growth increases continued through the fifth year.

In the present study, weed infestation was much higher on PRP 2 and sporadically influenced natural regeneration. The overall amount of seedlings reached 67,450 per ha (Table 3). The natural regeneration of Norway spruce prevailed (44.1%) even though the number of Douglas fir seedlings was only slightly lower (37.8%). The other tree species accounted for 18.1%.

On PRP 3, the situation was similar to that in 2006. Weed infestation was very strong. Weeds predominated in some places and thus enabled natural regeneration to grow. Some parts of PRP 3 were totally without seedlings. In total, 39,675 seedlings per ha were counted on the plot (Table 3). Douglas fir was the most frequent (54.4% of seedlings), followed by Norway spruce (38.9%) and by other tree species (6.7%).

The frequency distribution of tree species among the particular PRPs was proved to be significantly different (χ^2 = 344.58, df = 8, P << 0.01). Similarly like in 2006, the number of Douglas fir seedlings was significantly variable on the particular plots (χ^2 = 22.73, df = 2, P << 0.01). Even though the share of Douglas fir seedlings was the lowest on PRP 1, the total amount was still the highest in the area treated with Velpar (1,066; i.e. 26,650 seedlings per ha). The second highest number of Douglas fir seedlings was counted on PRP 2 (1,019; i.e. 25,475 seedlings per ha). Conversely to the first plot, the total number of Douglas fir seedlings was the lowest on PRP 3 (864; i.e. 21,600 seedlings per ha) but the share of the seedlings in comparison with the other species, including Norway spruce, was the highest on this plot.

The high seedling numbers counted during both seasons (2006 and 2008) show a high potential of natural regeneration. The chemical soil preparation using selective herbicides seems to be hardly replaceable. Graham et al. (1989) stated on the basis of their research that Douglas fir trees growing in bedded soils treated with herbicide were heavier, taller and had deeper root systems than trees growing in other preparations. Velpar extended the best effects during the present research. The active substance degraded fluently and operated in soil for several years. However, the active substance resistance in soil has both positive and negative consequences. It can negatively affect seedlings that are mostly vulnerable in the first years of development. The positive impact can be seen in the weed growth inhibition. Weed suppression was less evident on PRPs 2 and 3, where Roundup Forte and Dominator were applied. The situation was the worst on PRP 3. The efficacy of the herbicide active substance plays an important role. The active substance is decomposed very quickly after contact with soil in the case of Roundup Forte and Dominator. Thus the newly emerged weeds are not inhibited.

The importance of the chemical soil preparation is shown by a comparison of the three PRPs with

Table 4. Seedling number on PRP without chemical soil preparation after growing season 2008

Tree species	Seedling age					Seedling number per plot	Seedling number per ha
	1	2	3	4	5		
DF	19	9	5	5	0	38	950
NS	29	11	13	9	4	66	1,650
SF	90	21	6	0	0	117	2,925
SP	0	0	0	0	0	0	0
Others	9	17	8	0	0	34	850
Σ per plot	147	58	32	14	4	255	
Σ per ha	3,675	1,450	800	350	100		6,375

PRP – permanent research plot, DF – Douglas fir, NS – Norway spruce, SF – silver fir, SP – Scots pine

Table 5. Seedling mortality between 2006–2008

PRP No.	Tree species	Seedling mortality (%)		
		3-year	4-year	5 and more year
1	DF	33.9	78.6	83.9
	NS	86.4	58.8	30.2
	SF	21.7	68.2	77.8
	SP	100.0	100.0	100.0
	others	25.0	0.0	0.0
2	DF	69.7	95.1	98.5
	NS	98.9	94.9	72.2
	SF	31.8	90.0	100.0
	SP	100.0	100.0	100.0
	others	85.7	100.0	100.0
3	DF	4.7	68.6	76.1
	NS	93.1	69.8	69.2
	SF	52.4	84.6	100.0
	SP	100.0	100.0	50.0
	others	100.0	100.0	0.0

PRP – permanent research plot, DF – Douglas fir, NS – Norway spruce, SF – silver fir, SP – Scots pine

the check plot. The amount of seedlings (Table 4) is much lower on the check plot without chemical preparation and reaches only 7.3% of the seedling number on the plot treated with Velpar. We can assume that this situation is caused by weed infestation on the check plot. Weeds inhibit the seedling rooting and growth. This study confirmed conclusions drawn by Wiloughby (1996) that the control of competitive vegetation is essential for the successful tree development. The statistical analysis also certified a high variability in seedling numbers between PRPs and the check plot (χ^2 = 1,119.08, df = 12, $P << 0.01$). The number of Douglas fir seedlings was significantly higher on plots treated with herbicides than on the check plot (χ^2 = 926.84, df = 3, $P << 0.01$). Stein (1999), who investigated the growth and survival of Douglas fir after herbicide and manual treatments and compared the obtained results with the control, published the same results. In his study, six years after treatment, the competitive vegetation cover was much greater on untreated that on treated plots.

Even though the survival of seedlings is quite high on plots treated with herbicides (Table 5), it must be said that the resistance of the active substance in soil, mainly on PRP 1, which was treated with Velpar, can cause the death of many seedlings. Nevertheless, the positive impacts of herbicide application exceeded the negative ones as stated also by Stein (1999), who found out that each treatment tested in his study improved the development and growth of Douglas fir. The other factor that could decrease the seedling survival in the present study is forest weeds. Conditions for the seedling survival become worse with gradual weed infestation on PRPs. Seedlings that do not develop fast enough to overgrow weeds suffer from the lack of light. The other elements that can play their role are precipitation and temperature fluctuations and damage caused by game. Game damage marks were found on all three PRPs.

CONCLUSION

The present research was focused on the study of natural regeneration of Douglas fir under the parent stand shelter. The parent stand 441D10 is one of the oldest and most productive stands of Douglas fir in mixture with our domestic tree species that can be found in the area of the Training Forest Enterprise in Kostelec nad Černými lesy. The chemical soil preparation against forest weeds was done on permanent research plots. By then, weeds inhibited seedling establishment and growth.

The most effective herbicide was Velpar, which was used on PRP 1. In total, 32,650 Douglas fir seedlings per ha were counted on the plot treated with Velpar in 2006 and 26,650 seedlings per ha in 2008. Conversely, Dominator proved low efficacy in the control of weeds. The competitive vegetation cover was very high and thus conditions for seedling emergence became worse. Altogether, 21,925 Douglas fir seedlings in 2006 and 21,600 seedlings per ha in 2008 were found on the Dominator-treated plot. However, seedling numbers found out on the plot without chemical treatment were even lower – only 950 Douglas fir seedlings per ha in 2008.

The most suitable areas for the chemical soil preparation seem to be stands without natural regeneration where the weed cover inhibits seedling growth. It is necessary to apply the herbicides during the Douglas fir seed year. Monitoring done in Aldašín confirmed that Velpar 90 WSP in a concentration of 3 kg in 400 l water per 1 ha was the most effective herbicide for long-term weed suppression. Nevertheless, the availability and concentration of herbicides must always be discussed with the herbicide producer or provider and must be in agreement with the substances recommended for forest management in the year of application.

The total seedling number exceeded 35,000 per ha on all research plots. Douglas fir accounted for more than 30% of the individuals counted after growing season 2008. The observed tree species composition and number is thus sufficient for further growth and development of the stand as far as the stand production and stability are concerned.

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Received for publication July 7, 2009
Accepted after corrections February 15, 2010

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