Growth of pedunculate oak (Quercus robur L.) seedlings on blend-substrates based on consolidated brackish sludge

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Abstract

Either in its pure form or as a base for a blend-substrate, brackish sludge can be used as a growth medium for trees. The objectives of the experiment were (1) to evaluate the quality of consolidated brackish sludge as a growth medium, (2) to assess the influence of substrate blending on tree growth (leaf area, tree height, biomass), and (3) to evaluate the need for substrate blending.

Germinated acorns of the same mother tree were planted in containers and grown under controlled conditions. The substrate consisted of pure brackish sludge or of brackish sludge blended with peat, sand or wood chips, in stages of 25 volume percentage.

Chemical analysis of the brackish sludge showed a neutral pH, a conductivity of 3.60 mS/cm and a sufficient amount of macro elements. The addition of sand contributed to an accelerated desalination of the substrate.

The treatments with 100 % sludge and 25 % peat yielded a leaf area, a tree height and a biomass which were among the top three growing results. The growing results of the treatments with 25% sand and 50% peat were consistently among the top five.

The blending of peat in brackish sludge had no influence on leaf area, tree height and biomass of seedlings. The use of wood chips or of more than 25 % sand resulted in a significant decrease of these three growth parameters.

Foliage analyses of the trees grown on a substrate containing more than 50% sand or wood chips revealed deficits in available nutrients. These deficits manifested themselves in a loss of leaf area, tree height and biomass production.

The nutritional status and the growth parameters of oak seedlings on brackish sludge were favourable. No influence of the salt and the heavy metal concentrations on growth was observed. Consolidated brackish sludge is a suitable growth medium for trees.

Keywords : *brackish, consolidated, growth, pedunculate oak, sludge*

1. Introduction

Sludge raised by dredging the Zeeschelde estuary downstream of Antwerpen (Belgium) is a source of micro and macro-elements (De Pauw en Poelman, 1994), but elevated amounts of heavy metals and salt limit the range of the uses brackish sludge can be put to. It is not suitable for application as a fertiliser in agriculture and horticulture.

The sludge is placed in lagoons where sand is separated from the sludge by gravitation. The sand fraction is subsequently used in public works (Cockaerts, 1993), but until now the consolidated dredging sludge has been dumped.

On the basis of an average embankment of 2.5 m to 3 m for a sludge disposal site it has been estimated that an area of 80 ha is needed annually in Flanders to dispose of the sludge (Dercon and Vanhaute 1994). Planting trees on these disposal sites would contribute to the extension of the forested area in Flanders.

This paper presents the results of a greenhouse experiment on the growth of oak seedlings and heavy metal uptake on brackish sludge mixed with different substrates. The aim was to establish whether brackish sludge is a suitable substrate for afforestation and whether blending increases the growth chances of trees on brackish sludge.

2. Material and Methods

Experiments were carried out in the greenhouse of the Laboratory of Forestry of the University of Gent, located 12 km south-east of Gent, Belgium. Pedunculate oak (*Quercus robur L.*) was selected because of the vague soil requirements of this species and the high sensitivity for heavy metals of the seedlings (Dixon 1988). Germinated acorns, all of the same mother tree, were selected on the basis of germ root length. At the end of March acorns with a germ root of more than 2 cm long were planted in perforated plastic containers with a content of 3,500 cm^3. The seedlings were monitored through two growing seasons, over 550 days.

Three substrates were evaluated as blends with pure brackish sludge : sand, peat and wood chips. These substrates were added in 25% steps by volume : 0 % (pure brackish sludge), 25 %, 50 %, 75 % and 100 %, with 10 replicates each. The 150 containers were placed in a closed greenhouse. Total watering during the 550 days amounted to 2,920mm, and was adapted to the water requirement.

As the plant goes through a stress situation after being planted, the first growing season cannot be considered representative. In our experiment height, leaf area and oven dry biomass of each plant were measured after two seasons in view of minimising the impact of plant stress on growth. After drying at 105°C to constant weight leaves were analysed for P, K, Ca, Mg, Na, Zn, Cd, B, Mn, Fe, Al and Cu. Concentrations of these elements were determined by ICP-AES after digestion with HNO_3, a method that is recommended by the Society for Waste Products of the Flemish Government (OVAM, 1996; Kalra & Maynard, 1991). Nitrogen was determined by the Kjeldalh method.

According to Dixon (1988) and Van Den Burg (1974, 1985, 1990) seedlings may contain heavy metal concentrations that strongly deviate from saplings and trees that are 5 years old or older. The interpretation of the foliage analyses is based on reference values for adult trees. Consequently the results should be considered directive.

The substrate was analysed for ash rest, pH-H_2O, soil nitrogen content and conductivity. For pure brackish sludge these parameters were also measured at the start of the experiment. Ash rest was determined after ashing at 550°C, pH-H_2O and conductivity were measured in a suspension of 10 g substrate and 50 ml distilled water. Soil nitrogen was also determined with the Kjeldahl method (Kalra & Maynard, 1991, Labrecque et al., 1995). For the chemical analyses a single mixed-sample per treatment was made from the leaves as well as from the substrate. For pure brackish sludge an average value and a standard deviation, based on 3 mixed-samples of 10 sub-samples each, were calculated.

The data obtained from the blending sequences were tested for normality with the Shapiro-Wilk-test after the removal of the outliers. The blending sequence of sand could not be transformed to a normal distribution. Data from the blending sequences of peat, wood and pure brackish sludge were mutually compared by means of parametric tests (on the 95 % level). The overall comparison, which included sand, was executed with the Kruskal-Wallis test. Statistical calculations were executed with the SPSS software package.

3. Results

3.1. Substrate characteristics

At the start of the experiment the chemical characteristics of the brackish sludge were : pH-H_2O 7.7 , Kjeldahl nitrogen 3,400 mg/kg, ash rest 79 %, and electrical conductivity 3.60 mS/cm. The results of the substrate analyses after the experiment are shown in Table 1. Within two growing seasons the pH decreased from 7.7 to 7.1 and the total nitrogen content decreased from 3,400 mg/kg to 2,436 mg/kg.

The pH-H_2O was neutral, and no differences could be found between the treatments .

The total nitrogen content rose with an increased blending with peat or wood. Increased proportions of sand decreased total nitrogen content. The ash rest decreased with increased peat and wood proportions, and slightly increased with an increased sand proportion. A decrease of electrical conductivity was observed with an increasing amount of sand.

Table 1. pH-H_2O, Kjeldahl nitrogen content, ash rest and electrical conductivity (EC) for the 13 treatments at the end of the second growing season

Treatment (volume %)	pH-H_2O (-)	Kjeldahl nitrogen (mg/kg)	Ash rest (%)	EC (after 550 days) (mS/cm)
Brackish sludge				
100	7.1 ± 0.1	2436 ± 96	85.3 ± 1.4	0.31 ± 0.06
Peat				
25	6.9	2904	81	0.53
50	6.9	3735	77	0.42
75	6.8	5327	71	0.61
100	6.7	6656	67	0.33
Sand				
25	6.9	1344	92	0.23
50	6.8	869	95	0.15
75	7.1	512	97	0.14
100	6.9	120	99	0.10
Wood chips				
25	7.1	2528	83	0.33
50	7.2	3240	73	0.49
75	7.0	3564	63	0.34
100	6.7	17107	11	0.37

3.2. Influence of substrate blending on the tree height

The biggest height increment was found in the treatments with 100% sludge, 25 % peat, and 25 % sand respectively (Fig. 1). Compared to the treatment with 100% sludge, the blends of 25%, 50%, 75%, and 100 % peat did not affect the tree height. After 550 days all treatments with wood chips proved to have a significantly lower tree height than the treatment with 100% sludge.

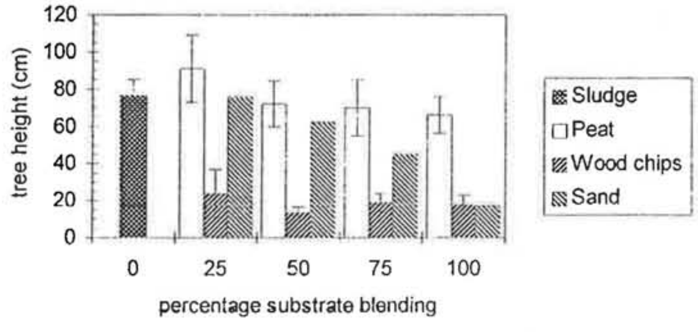

Figure. 1. Tree height of pedunculate oak seedlings for the different treatments after 550 days.

3.3. Influence of substrate blending on the leaf area

The treatments with 100 % sludge, 25 % peat, and 25 % sand yielded the largest leaf area (Fig. 2). Compared to the treatment with 100% sludge, the blends of 25 %, 50 %, 75 %, and 100 % peat did not affect the leaf area. The treatments with 25 % or more wood yielded a significantly smaller leaf area than the treatment with 100 % sludge.

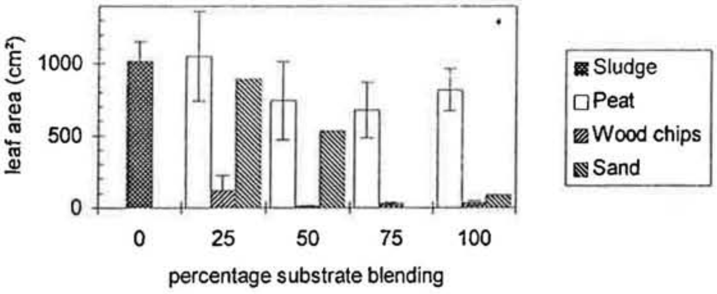

Figure 2. Leaf area of pedunculate oak seedlings for the different treatments after 550 days.

3.4. Influence of substrate blending on the biomass

The treatments with 100 % sludge, 25 % peat, and 25 % sand yielded the highest biomass (Fig. 3). The blends of 25%, 50%, 75%, and 100 % peat did not affect the biomass. The treatments with 25 % or more wood yielded a significantly lower biomass than the treatment with 100 % sludge.

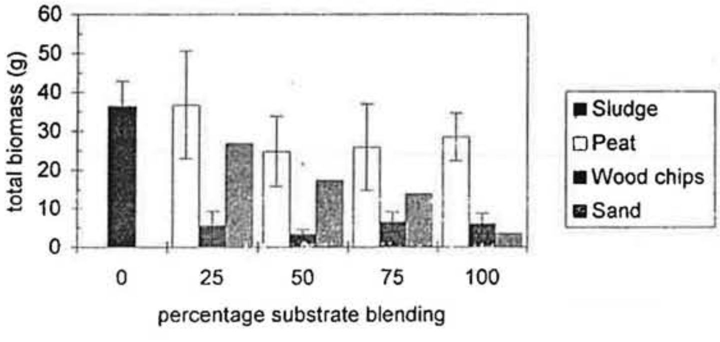

Figure 3. Total biomass of the pedunculate oak seedlings for the different treatments after 550 days.

4. Foliage analysis

The blending sequence of pure brackish sludge and peat contained a sufficient to optimal amount of plant available nutrients (Table 2). The nitrogen content of the leaf reached a deficit with an increasing volume percentage of wood and sand. An increase of the peat volume in the blend did not significantly change the nitrogen content in the leaf.

The phosphorus content reached a deficit in blends with 50 % wood and was low in the 100 % peat substrate. All other blends had a sufficient to optimal phosphorus content. Potassium, calcium, and magnesia contents reached a deficit when the substrate contained 50% wood chips. Other blends produced optimal leaf content of these elements.

For the blending sequence with wood the amounts of leaves were insufficient for analysis when the substrate contained 75 % to 100 % wood. The treatments with 50 % wood showed a deficit for all analysed elements. For the blending sequence with sand a deficit for nitrogen content was observed.

In the blending sequence of sand a Pearson correlation coefficient of 0.98 was found between the nitrogen content in the soil and the nitrogen content in the leaf. This correlation was not found in the other sequences.

Table 2. Nutrients contents in the leaf, expressed in mg/kg dry weight; ^{o} = optimal, ^{s} = sufficient, ^{l} = low and ^{d} = deficit (Van Den Burg, 1985), n/a = no sample available

Treatment (volume %)	N	P	K	Ca	Mg	Na*
Brackish sludge						
100	30735$^{o/s}$ ± 5883	2567$^{o/s}$ ± 613	10442^{o} ± 1714	18614^{o} ± 4857	4572^{o} ± 533	961 ± 739
Peat						
25	24658^{s}	2413^{o}	10550^{o}	20791^{o}	4489^{o}	1066
50	32542^{o}	2498^{o}	11225^{o}	21898^{o}	5146^{o}	1038
75	26480^{s}	2167^{o}	10150^{o}	18309^{o}	4582^{o}	1136
100	29877^{o}	1038^{l}	9750^{o}	17477^{o}	3181^{o}	1141
Sand						
25	22110^{s}	2201^{o}	9750^{o}	23023^{o}	6443^{o}	879
50	12198^{d}	2471^{o}	9900^{o}	20909^{o}	4876^{o}	787
75	8954^{d}	2542^{o}	9450^{o}	21382^{o}	4231^{o}	1127
100	6900^{d}	1669^{s}	8300^{o}	20533^{o}	4237^{o}	1092
Wood chips						
25	23700^{s}	2144^{o}	9400^{o}	22507^{o}	5072^{o}	841
50	9212^{d}	2178^{d}	1250^{d}	6848^{d}	605^{d}	313
75	n/a	n/a	n/a	n/a	n/a	n/a
100	n/a	n/a	n/a	n/a	n/a	n/a

* Van Den Burg gives no reference values for Na

Table 3 shows an accumulation for Zn for both pure brackish sludge and the peat and sand sequences. The manganese concentration in the leaf was low and caused a deficit in the seedlings on pure brackish sludge. Other trace elements were present in high, normal or low concentrations for pure sludge and the peat and sand sequences. The blending sequence with wood chips shows a Cu deficit, while the other trace elements were present in high, normal or low concentrations (Table 3).

Table 3. Contents of trace elements in the leaf, expressed in mg/kg dry weight; ^{a} = accumulated, ^{h} = high, ^{n} = normal, ^{l} = low, ^{d} = deficit (Heinrichs & Mayer, 1980; Martin & Coughtrey, 1981), n/a = no sample available

Treatment (volume %)	Zn	Cd	B	Mn	Fe	Al	Cu
Brackish sludge							
100	$343^{h/a}$	2.5^{h}	78^{n}	$37^{d/l}$	531^{h}	$82^{n/h}$	5.4^{n}
	± 164	± 0.3	± 10	± 25	± 282	± 16	± 2.7
Peat							
25	418^{a}	3.2^{h}	72^{n}	41^{l}	525^{h}	68^{n}	4.9^{n}
50	413^{a}	2.2^{h}	66^{n}	49^{l}	563^{h}	87^{n}	6.1^{n}
75	317^{h}	2.5^{h}	71^{n}	36^{l}	386^{h}	53^{n}	5.4^{n}
100	401^{a}	2.6^{h}	131^{h}	251^{n}	545^{h}	78^{n}	5.3^{n}
Sand							
25	353^{h}	2.0^{h}	84^{n}	44^{l}	544^{h}	145^{h}	5.6^{n}
50	348^{h}	2.9^{h}	58^{n}	58^{l}	486^{h}	84^{n}	4.5^{n}
75	402^{a}	2.6^{h}	67^{n}	60^{l}	583^{h}	102^{h}	4.8^{n}
100	278^{h}	2.2^{h}	54^{n}	66^{l}	372^{h}	56^{n}	5.5^{n}
Wood chips							
25	330^{h}	3.3^{h}	87.4^{n}	114^{l}	568^{h}	164^{h}	6.4^{n}
50	89^{n}	2.6^{h}	25.8^{n}	77^{l}	163^{n}	46^{n}	1.7^{d}
75	n/a	n/a	N/a	n/a	n/a	n/a	n/a
100	n/a	n/a	N/a	n/a	n/a	n/a	n/a

5. Discussion

5.1. Substrate characteristics

The ionic strength in pure brackish sludge was 3.60 mS/cm at the start of the experiment. The fast washout of minimum $5.4\ 10^{-3}$ mS/ cm.day and maximum $6.4\ 10^{-3}$ mS/ cm.day is probably due to the high amount of water used in irrigation and the use of separated sludge. The blending sequence of sand has a significantly lower electrical conductivity than the other blending sequences . This indicates that for the afforestation of sludge, the blending with sand can lead to an accelerated decrease of salt stress.

The low total nitrogen content for the blending sequence with sand is normally explained by the low organic nitrogen content, illustrated by the high percentage of ash rest (Table 1). The ash rest, which is a measure of the content of non volatile, inorganic elements (OVAM 1996), is the highest for the sand substrate and the lowest for the wood substrate . Overall, the total nitrogen content is strongly but negative correlated with the ash rest : Pearson correlation coefficient of -0.97.

5.2.Tree height, leaf area and biomass

A pot experiment with oak seedlings planted in sand-loam soil and placed under a natural cover resulted in a tree height of 22.3 cm after one growing season (Lust & Speleers, 1990). A similar set-up without a cover yielded a tree height of 17.3 cm (Lust & Speleers, 1990). A pot experiment with a substrate of 20 % clay, 20 % fine sand, and 60 % peat, yielded a tree height of 36 cm in the open field, and 31 cm under an artificial cover after one growing season (Van Hees, 1997). Oak seedlings planted in brackish sludge yielded an average tree height of 30 cm after one growing season. After one growing season the tree height was 7.7 cm to 12.7 cm higher on brackish sludge than on sand-loam. The tree height was 6 cm, or 20 %, lower on brackish sludge than on an optimal clay-sand-peat substrate (Van Hees, 1997).

As the leaf area is determined by the gas and energy exchange between the plant and its environment, it provides a picture of the productivity (McIntrye et al. 1991, Wang et al. 1992). Pot experiments with pedunculate oak, planted in a sand-loam substrate, yielded a leaf area of 42.5 cm^2 after one growing season under cover. A leaf area of 47.2 cm^2 was obtained when the seedlings were grown in the open field (Lust & Speleers, 1990). On the contrary the leaf area in the pot experiment with blend-substrates based on brackish sludge amounted to an average of 987 cm^2 for the three highest blends after two growing seasons.

In accordance with the OECD guidelines for biological toxicity tests (1984) biomass was also defined as a growth parameter. Lust & Speleers (1990) report a total biomass 0.803 g for pedunculate oak under cover. Pedunculate oak grown in the open field reached a biomass of 0.8022 g after one growing season. Van Hees from his side (1997) obtained a biomass of 17 g under an artificial cover. In the open field a biomass of 34 g was obtained after one growing season. The average biomass for seedlings grown on brackish sludge was 36.4 g after two growing seasons.

5.3. Foliage analysis

The content of nutrients in the leaf provides an indication for the amount of plant available nutrients in the substrate (Van Den Burg 1974, 1985, 1990). The total amounts of nutrients appeared not to be different for wood and for peat and brackish sludge. The chemical form in which these nutrients are present may explain the lesser growth on wood chips.

The nitrogen in the blending sequence with sand is present in a plant available form. In the sand blend the amount of plant available nitrogen is practically equal to the total amount of nitrogen. This is illustrated by the strong correlation between the nitrogen content in the soil and that in the leaf. In the other blending sequences part of the nitrogen is available as organic material. This part of the total nitrogen content is not plant available and is not found in the leaf. There is no correlation between the nitrogen content in the soil and that in the leaf.

6. Conclusions

The nutritious status and the growth parameters of oak seedlings on brackish sludge are favourable. The salt and heavy metal concentrations had no observable influence on growth. Consolidated brackish sludge is a suitable growth medium for trees.

The results of the treatments with 100 % sludge and 25 % peat are among the top three for tree height as well as for leaf area and biomass. The treatments with 25% sand and 50% peat yield results that are consistently among the top five. The blending of peat with brackish sludge has no impact on biomass, tree height and leaf area of pedunculate oak seedlings . The use of either wood chips or more than 25 % sand results in a significant decrease of these three parameters.

The foliage analysis shows that there are shortages in plant available nutrients in a growth medium that consists of more than 50 % sand or wood chips. This is manifested in a loss of tree height, leaf area and biomass production. No correlation was found between the concentrations of heavy metals in the leaf and the growth parameters.

Although the growth results of brackish sludge blended with sand are less favourable than those of pure brackish sludge, the blending of sand could be considered for large-scale afforestation on consolidated strong brackish sludge. In an optimal proportion sand contributes to an accelerated desalination, subject to some loss in growth. Accelerated desalination reduces the risk of salt stress for the trees. Setting up these disposal sites in the immediate vicinity of brackish rivers is advisable in order to avoid the salting of the ground water.

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8. References

Cockaerts, G. & Dekeyzer, C. (1993). Proefprojekt Kallo, revalorisatie van baggerspecie uit de Beneden-Zeeschelde. Ondernemingen Jan De Nul in samenwerking met Laboratorium voor Bosbouw (RUG), deelrapport. 73 p.

De Pauw, N. & Poelman, E. (1994). Waterverontreiniging. *In*Leren om te keren Milieu en natuurrapport Vlaanderen. Ed. A Verbruggen. pp 319-347. Vlaamse milieumaatschappij, Garant. Leuven, Apeldoorn.

Decron, L. & Vanhaute L. (1994). Infrastructuren. *In*Leren om te keren Milieu en natuurrapport Vlaanderen. Ed. A Verbruggen. pp 167-183. Vlaamse milieumaatschappij, Garant. Leuven, Apeldoorn.

Dixon, R. (1988). Response of ectomycorrhizal Quercus rubra to soil cadmium, nickel and lead. Soil Biology and Biochemistery **20**, 555-559.

Heinrichs, H. & Mayer, R. (1980). The role of forest vegetation in the biological cycle of heavy metals. Journal of Environmental Quality **9**, 111-118.

Kalra, Y. P. & Maynard, D. G. (1991). Methods for forest soil and plant analysis. Information report NOR-X-319, Forestry Canada, Northwest Region, Northern Forestry Centre. 116 p.

Labrecque, M., Teodorescu, T. & Daigle, S. (1995). Effects of wastewater sludge on growth and heavy metal bioaccumulation of two *Salix* Species. Plant and Soil **171**, 303-316.

Lust ,N. & Speleers, L. (1990). The establishment of red oak and pedunculate oak seedlings in the experimental forest of Aelmoeseneie at Gontrode (Belgium). Silva-Gandavensis **55**, 1-23.

Martin, M. H. & Coughtrey, P. (1981). Impact of heavy metals on ecosystem function and productivity. *In*The effects of heavy metal pollution on plants vol.2. Ed. N Lepp. pp 119-158. Applied Science Publishers, London and New York.

Mclntrye, B. M., Scholle, M. A. & Sigmon, J.T.A. (1991). A quantitative description of a deciduous forest canopy using a photographic technique. Forest Science **36**, 381-393.

OCDE (1984). OECD Guidelines for testing of chemicals. Organisation for Economic Cooperation and Development, Paris.

OVAM (1996). Afvalstoffenanalyse compendium. OVAM, Mechelen.

Van Den Burg, J. (1974). Toepassing van de bladanalyse bij jonge loofhoutopstanden. De Dorschkamp, Wageningen. 615 p.

Van Den Burg, J. (1985). Foliar analysis for determination of tree nutrient status - a compilation of literature data. rapport 414. De Dorschkamp, Wageningen. 468p.

Van Den Burg, J. (1990). Foliar analysis for determination of tree nutrient status a compilation of literature data rapport 591. De Dorschkamp, Wageningen. 220 p.

Van Hees, A. F. M. (1997). Growth and morphology of pedunculate oak (*Quercus robur* L) and beech (*Fagus sylvatica* L) seedlings in relation to shading and drought. Annales des sciences forestières **54**, 9-18.

Wang, Y. S., Miller, D. R., Welles, J.M. & Heisler, G.M., et al. (1992). Spatial variability of canopy in an Oak forest estimated with fisheye sensors. Forest Sciencc **33**, 210-216.