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Original Research

Relations between the Yield of Pine and Ca/Al Ratio in the Growth Environment and Plants

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Abstract

Relations between the yields of pine and Ca/Al ratio were studied in two experiments. In the first experiment one-year-old pine seedlings were grown in soil-water cultures. The source of elements was the soil from O, AE and Bv horizons sampled under pine stands. In the second experiment two-year-old pines were cultivated in quartz sand and increasing doses of Al were added. The results showed that the yield of one-year-old pine was not related to the Ca/Al value, neither in soil solution nor in plants. Increasing doses of Al from 2 to 100 mg Al kg^{-1} decreased the yield from 42 g to 13 g d.w. and Ca/Al values from 8.1 to 1.0 in needles, from 4.4 to 1.0 in shoots and from 0.63 to 0.07 in roots. The correlation coefficient between Ca/Al and yield of needles was 0.850, between Ca/Al and yield of shoots was 0.867 and between Ca/Al and yield of roots was 0.712.

Keywords: Al, pine, yields, Ca/Al ratio

Introduction

The mechanism of Al toxicity in various plant species has not been conclusively recognized [1]. Kochian et al. [2] summarizing results from a number of studies pointed out that plant resistance to Al depends on physiological and genetic properties of plants. According to Zheng et al. [3], Al in the cell wall is easily bound mainly by pectates, but at the same time it is easily replaced by Ca. Jozefaciuk and Szatanik-Kloc [4] associate Al toxicity with the quantity of cations adsorbed by plant roots. The authors suggest that cation sorption in roots depends on the size and density of negative charge on the root surface. Kikui et al. [5] suggest that Al tolerance in wheat depends on the Al-triggered efflux of malate from root apices. According to other authors, Al stress in plants is to be observed as an impairment of metabolic processes [6-8].

Symeonidis et al. [9] provided evidence of a strong response of *Cucumis melo* to Al exposure when its content in the medium was 0.2 mmol L^{-1}, a decrease in leaf area, chlorophyll content, plant biomass and Ca content in plants were observed. Ghanati et al. [10] on the other hand, observed the increased growth of tea plants induced by Al. Likewise, Watanabe and Osaki [11] found an increase in the growth of *Melastoma malabathricum*.

Some authors have concluded that Al toxicity in plants can be alleviated in the presence of Ca and other base cations when their plant-available content in the growth environment exceeds that of Al [12-14]. Al interferes with Ca uptake and translocation from roots to aboveground plant parts. Therefore, attention was turned to the question whether and to what extent the Ca/Al ratio in soil and soil solution, in the medium or in plants may constitute a complex indicator of Al toxicity to plant growth. Authors suggest that Al toxicity may be measured by a decline in the growth of plant roots [15, 16].

It has been found that the molar Ca/Al ratio in the soil solution provides a valid indicator for the evaluation of acidic degradation of soils [17-21]. Molar Ca/Al ratios in foliage and plant roots are also regarded as indicators of Al toxicity to plants, including forest plants [15, 16, 22-24].

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In the majority of studies the indicator value was assessed on the basis of plant root growth and on the basis of morphological changes in roots subject to Al stress [15, 24]. Hansen et al. [25] studied the (Ca+Mg+K)/Al and Ca/Al ratios in soil solution under spruce and beach in relation to vitality and health stands. Authors questioned the suitability these ratios as the chemical criteria for soil acidification under field conditions, but stated that constantly decreasing these ratios would cause forest damage. Similarly Hruska et al. [26] could not set strict criteria for forest dieback due to a decline of Ca/Al or BC/Al molar soil solution ratios. Furthermore, Nygaard and De Wit [27] stated that the key role of Al toxicity, expressed as the Ca/Al ratio in critical load calculations for forest was insufficient.

In this study an attempt was made to establish:

- relationships between the Ca/Al ratios in the soil, soil solution, pine roots and aboveground parts and the amount of biomass produced;
- effect of increasing doses of Al on the yields and relationships between the yields and Ca/Al ratio in pine needles, shoots and roots.

Materials and Methods

In the first experiment one-year-old pine seedlings were cultivated in soil-water cultures in a greenhouse, in 3 L pots having 20 cm in diameter. Pine seeds were sown in quartz sand and, after the roots appeared, the seedlings were mounted on stands installed in pots. To ensure proper aeration the plant roots remained above the soil solution to one third of their length. The sources of elements were soils from pine stands sampled from individual genetic horizons of the main rooting layer. Rustic Podzol [28, 29], was represented by three genetic horizons, i.e. O, AE and Bv. The pots were filled in with 0.2 kg of soil from the O horizon and 1 kg of soil from AE and Bv horizons, respectively, and poured with 2.5 L of deionized water. The plants (30 plants per pot) were cultivated from May through October. In the experiment extra variants were introduced, i.e. to the soils of AE and Bv horizons, N was added in the amount of 80 mg N kg^{-1} as NH_4NO_3.

The solution above the soil in pots was supplemented by deionized water to constant volume every week. The content of Ca and Al was determined five-fold (once a month) throughout the vegetation season, i.e. the 100 mL of solutions were taken from each pot and filtered using the cellulose filter (2 μm), and next Ca and Al were determined using the FAAS method.

Before the onset of the experiment the soils were analyzed [30] for: organic C using Tiurin's method and total N using Kiejdahl's method, pH in H_2O (1:2.5 mass to volume ratio), exchangeable cations (Ca, Mg, K, Na) after extraction with 1M ammonium acetate (pH 7), exchangeable Al and H after extraction with 1 M KCl (1:10 mass to volume ratio). In all extracts elements were determined using the FAAS method.

In the second experiment two-year-old pine were cultivated on quartz sand (pots 30 cm diameter). Pine seeds were sown to the hotbed in May one year before, and in May next year five individual plants (mass of each plant was about one gram f.w.) from the hotbed were put in each pot. The following nutrients were applied: N – 80 mg kg^{-1} as NH_4NO_3, P – 20 mg kg^{-1} as KH_2PO_4, K – 60 mg kg^{-1} as KH_2PO_4 and KCl, Ca – 50 mg kg^{-1} as $CaCl_2$, Mg – 20 mg kg^{-1} as $MgSO_4$. Al was applied as $AlCl_3$ at increased doses from 2 to 200 mg kg^{-1}. The fertilizers and $AlCl_3$ were mixed with 5 kg of sand before filling the pots. Plants were grown from May through October, and pots were watered to a constant weight every day in order to maintain the moisture degree at a level of 10% of weight, which is equivalent to the field water capacity of the sand.

Upon termination of the growing season, plants were dried up (after rinsing with deionized water), weighed and analyzed. Ca and Al were determined in mineralized samples using the MDS-2000 microwave digestor by FAAS method.

Both experiments were made in three replicates. The results were given as an arithmetical mean of three replicates. Differences between values obtained for both the yield quantity and element content did not exceed 10%.

Additionally, to compare the relationship between the yield and Ca/Al ratio in pine growing in laboratory and field conditions, the one-year-old pine seedlings were taken in September from seven commercial nurseries. In each nursery 200 individual seedlings were collected from the area of 1000 m^2. After rinsing with deionized water the length of roots and shoots were measured and plants were dried and analyzed as in the other experiment.

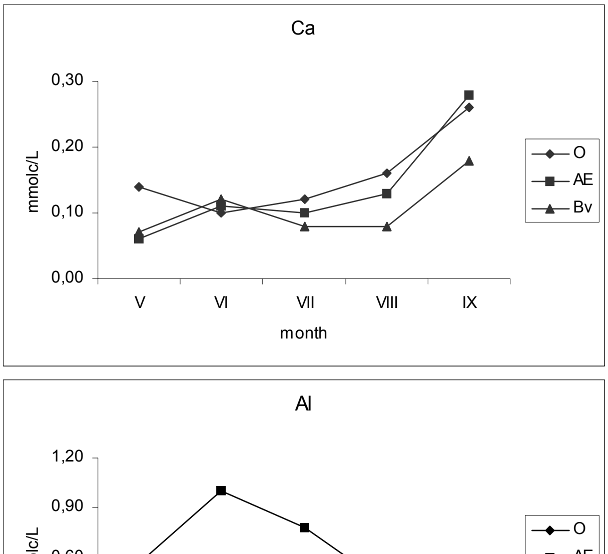

Fig. 1. Ca and Al dynamics in soil solution during pine growth on rusty podzolic soil.

Table 1. Some characteristics of soils used.

Soil	Horizon	C_{org}	N_{tot}	pH_{H_2O}	Exchangeable, $mmol_c$ kg^{-1}			< 20 µm
		%			Ca+Mg+K+Na	Ca	Al	
rusty podzol	O	43.0	1.300	3.7	122.7	90	26.0	
	AE	3.04	0.107	3.7	4.9	3.0	37.2	4
	Bv	0.50	0.025	4.5	1.0	0.4	12.3	6

Table 2. Yield and Ca/Al ratio in soil, soil solution and pine.

Horizon	Soil kg pot^{-1}	Yield* g d.w.	Soil, mmol kg^{-1}			Soil solution, mmol L^{-1}			Shoots, mmol kg^{-1}			Roots, mmol kg^{-1}		
			Ca	Al	Ca/Al	Ca	Al	Ca/Al	Ca	Al	Ca/Al	Ca	Al	Ca/Al
O	0.2	3.7	45	8.7	5.17	0.08	0.12	0.67	39.0	9.7	4.02	15.5	26.0	0.60
AE	1	6.8	1.5	12.4	0.12	0.07	0.17	0.41	25.0	24.7	1.01	3.6	77.7	0.05
AE+N	1	8.2	1.6	12.6	0.13	0.06	0.23	0.26	27.5	22.1	1.24	5.0	59.3	0.08
Bv	1	4.4	0.2	4.1	0.05	0.06	0.10	0.60	17.5	18.0	0.97	5.0	166.7	0.03
Bv+N	1	8.3	0.2	3.7	0.05	0.06	0.08	0.75	18.0	22.3	0.82	5.0	159.3	0.03

* mass of 30 plants

Ca/Al - molar ratio; contents of Ca and Al in soil solution mean values of 5 measurements

source of N - NH_4NO_3

Results

Rusty podzol soil used in the first experiment was strongly acidic and in AE and Bv horizons contained large amounts of exchangeable Al and low content of Ca (Table 1). The concentrations of Al in the solutions were higher than those of Ca. The Ca content in solutions indicated a growing trend over the entire vegetation season. Throughout the first month of the vegetation season the Al content in soil solutions increased while in the following months it decreased markedly (Fig. 1).

Molar ratios of exchangeable Ca and Al in soil horizons were: O – 5.17, AE – 0.12 and Bv – 0.05, respectively. The solutions that were in balance with the soil throughout the vegetation season were found to have the following Ca/Al ratios (five measurements average) in the respective horizons: O – 0.67, AE – 0.41 and Bv – 0.60 (Table 2).

The Ca content in shoots ranged from 17.5 to 39 mmol kg^{-1} while in roots from 3.6 to 15.5 mmol kg^{-1}. No relationship was found between Ca content in the soil solution and shoots or roots (Table 2).

The Al content in the aboveground parts of pine ranged from 9.7 to 24.7 mmol kg^{-1} while in the roots - from 26 to 166.7 mmol kg^{-1}. The Ca/Al ratio in the shoots of pine varied depending on genetic horizons of the soils applied from 0.82 to 4.02 and the lowest value was found for Bv horizon with N treatment. The root Ca/Al ratio was lower than 0.1, except O horizon (0.60). The plant yields ranged from 3.7 to 8.3 g, and the highest yields were obtained where N treatment was applied. The yield was not related to the Ca/Al ratio in the soil, above-soil solution or in plants (Table 2).

The low doses of Al (2 and 10 mg kg^{-1}) had no impact on biomass, though they decreased the Ca/Al ratio in plants. The 2 mg kg^{-1} dose decreased the Ca/Al ratio only in the roots of pine, while the dose of 10 mg kg^{-1} had similar effect for the whole plant. Increasing Al doses from 20 to 200 mg kg^{-1} resulted in decreasing yields ending up with the dieback of plants at the highest dose. The increase in Al dose was associated with increasing Al and decreasing Ca contents and, as a result, in decreasing Ca/Al ratio in plants. Under conditions of this experiment a strict relationship was found between the yield and the Ca/Al ratio in needles, shoots and roots (Table 3).

The average lengths of both shoots and roots of one year-old pine from seven commercial nurseries were similar, whereas Ca/Al ratios ranged from 3.67 to 0.86 in shoots and from 0.87 to 0.14 in roots (Table 4). No relationship was observed between the length of shoots and roots and the Ca/Al ratio in these parts of pine (correlation coefficient respectively 0.154 and 0.312).

Discussion

We found that Al content in the soil solutions constituted from 1.4% for O horizon to 2.4% for Bv horizon of its exchangeable pool, whereas Ca constituted from 0.2% for O horizon to 30% for Bv horizon of its exchangeable pool. These results are in agreement with earlier investigations concerning the evaluation of interrelationships between the pool of exchangeable cations in the soil and their pool migrating to soil solutions [31]. However, exchangeable Ca

Table 3. Effect of Al stress on pine yields and Ca/Al ratio.

Al.	Yield*	Needles			Shoots			Roots		
		Ca	Al	Ca/Al	Ca	Al	Ca/Al	Ca	Al	Ca/Al
mg kg^{-1}	g d.m.	mmol kg^{-1} d.m.			mmol kg^{-1} d.m.			mmol kg^{-1} d.m.		
0	42.5 ± 3.0	90.0	14.8	6.08	70.0	14.8	4.73	57.5	46.3	1.24
2	42.2 ± 2.9	120.0	14.8	8.11	81.3	18.5	4.39	52.5	83.3	0.63
10	41.3 ± 3.2	90.0	24.0	3.75	75.0	26.0	2.88	42.5	92.6	0.46
20	32.1 ± 3.1	68.8	24.0	2.87	65.0	26.0	2.50	35.0	92.6	0.38
40	22.2 ± 2.9	65.0	29.6	2.20	57.5	29.6	1.94	28.8	107.4	0.27
100	13.5 ± 2.0	50.0	50.0	1.00	41.3	40.7	1.01	11.3	152.0	0.07
200	2.6 ± 1.8	47.5	60.9	0.78	36.3	45.7	0.79	10.0	148.1	0.07

* mass of 5 plants

correlation coefficient between the yields and Ca/Al ratio: in the needles 0.850, shoots 0.867, roots 0.712

Table 4. Growth and Ca/Al ratio in biomas of one-year-old pine in commercial nurseries.

Commercial nurseries	Mass of 100 plants	Part of plants	Length	Ca	Al	Ca/Al
	g d.w.		cm	mmol kg^{-1} d.m.		
Dobrzany	54.2	shoots	6.8	6.8	2.1	3.24
		roots	16.3	5.8	8.2	0.71
Lubiechowo	59.0	shoots	5.7	4.3	1.8	2.39
		roots	17.0	2.0	5.2	0.38
Mirosławiec	66.0	shoots	5.6	2.8	1.8	1.56
		roots	17.0	2.7	3.3	0.82
Bobrowice	92.0	shoots	6.1	5.5	3.3	1.67
		roots	19.0	3.3	8.1	0.41
Kolonowskie	52.0	shoots	6.0	2.5	2.9	0.86
		roots	16.0	1.0	7.0	0.14
Grochowo	32.3	shoots	4.9	8.8	2.4	3.67
		roots	15.1	5.2	6.0	0.87
Tuchola	96.0	shoots	5.8	5.4	4.8	1.13
		roots	16.5	2.5	3.1	0.81

correlation coefficient between: length of: shoots and Ca/Al 0.154, length of roots and Ca/Al 0.312; Ca/Al shoots and Ca/Al roots 0.454

in O horizon was distinctly higher than in mineral horizons, but then in soil solution differences between Ca content in organic and mineral horizons were low. The highest concentration of Al was found in soil as well as in soil solution for AE horizon.

The Al saturation of forest soil sorption complex often attains more than 90%, thus the amount of basic cations is low [32, 33]. The speed of migration to soil solution was found to be higher for Ca than for Al ions, nonetheless, in soil solutions, frequently there is more Al than Ca ions [31].

In our experiment the change of concentration of both Ca and Al in soil solution during the vegetation of pine was observed. For example, for AE horizon we found the concentration of Al higher than 1 mmol$_c$ L^{-1}, especially in the second month of vegetation, whereas for Bv horizon only about 0.4 mmol$_c$ L^{-1}. At the end of the vegetation period the content of Al in soil solution for Bv horizon was very low. Similar data obtained by Patel-Sorrentino et al. [34] in infiltered soil water from an upper layer of podzolic soil, whereas Kim et al. [35] in water extracts from A horizons found higher Al concentrations.

We found that the Ca/Al values in pine plants depend on soil properties. In the shoots the Ca/Al value fluctuated from 1.01 to 4.02 in variants with O and AE horizons, and was < 1 in Bv horizon. The Ca/Al value in the roots, when soil of O horizon was applied, was 0.60, while for AE and Bv horizons they were lower than 0.1. No relationship was found between biomass yield and values of Ca/Al in plants, nor between values of Ca/Al in the above-soil solutions and Ca/Al in plants. Results of Eldhuset et al. [36] indicate that the reaction of root biomas and the Ca/Al ratio in the roots of *Picea abies* stand for Al concentration in the soil solution up to 0.2 mmol L^{-1} is not clear. Similarly De Wit et al. [37] stated that potentially toxic Al concentration in soil solution did not affect forest vitality and fine roots of *Picea abies* growths, but a decrease of Mg/Al ratio in the roots was observed.

Bakker et al. [22] reported the Ca/Al ratio within the range of 0.1 0.2 in fine root oaks sampled from four diverse depths in both the first year after the soil was limed and after a lapse of 27 years since the time of lime amendment was made.

Camps Arbestain et al. [21] have found that the ratio of water-soluble Ca/Al in the top layer (A horizon) of soils developed of granite was about 0.2, while in the soils developed of amphiboles - 5.82, whereas in fine roots of woody species sampled from the above soils, the Ca/Al ratio was 0.35 and 0.13, respectively.

Fernandez-Sanjurjo et al. [38] found that Al is even in as much as in 85% complexed by the organic matter in the organic and mineral-organic horizons of acid forest soils and with a predominance of monomeric Al forms in soil solutions. Yagasaki et al. [39] suggest that the activity of Al^{3+} for A horizons are controlled by complexation with soil organic matter and for B horizons by short-range ordered aluminosilicates. Van Hees et al. [40] found that Al in soil solutions occured mainly in monomeric forms, which according to Kopittke et al. [41] are much less toxic than polymeric compounds.

Our results are coincident with results of other authors concerning the low toxicity of Al in soil solution for pine. The differentiation of Al content in plants, and particularly in plant roots, may be associated with the Al content in the soil solution and with the properties of soil genetic horizon. The uptake of Ca and Al by pine is controlled by soil properties, including the soil solution content of Ca and Al.

In our second experiment, it was found that when applying increasing Al doses the pine reacted by decreasing the Ca/Al ratio. While the concentration of Al was 20 mg kg^{-1} or higher successive decrease in yields was observed. The decrease in Ca/Al ratio, especially in roots resulting from both the increasing accumulation of Al and decreasing accumulation of Ca. Under conditions of this experiment a strict relation was observed between yield and Ca/Al ratio in pine.

Summarizing papers of many authors Cronan and Grigal [15] stated that there was a correlation between Ca/Al ratio in the medium and Ca/Al ratio in spruce needles. Decreasing the Ca/Al ratio in the medium caused a decrease of Ca/Al in the needles. Ca/Al ratio of 0.2 in roots of forest woody plants accounts for the Al toxicity risk of 50%, or the impairment of plant roots, while at the Ca/Al ratio of 0.1 the risk increases to 80%. Our results are coincident with Cronan and Grigal [15] results when increasing doses of Al were applied with stable concentration of other ions.

Ca/Al ratio in pine needles is generally lower than 6.5, and even lower than 1.8 were found by Alvarez et al. [16], and the latter value was assumed by Truman [after 16] to be critical for *Pinus taeda*. At the same time no negative changes in stand growth were discovered. The Ca/Al ratio in pine needles (n = 280) was found to be about what Ostrowska et al. [42] explained by referring to the extreme soil acidity of sites where the relevant pine stands have grown (pH in H_2O was 3.5-4.5).

Some authors suggest that Al toxicity may be measured by the decline in growth of plant roots [15, 16]. In our observation no relationship was found between the length of roots and shoots and Ca/Al ratio in pine from commercial nurseries. We agree with Nygaard and De Wit [27] that the discrepancy between responses to Al exposure of tree seedlings in laboratory studies and trees from woodland sites suggest that the Ca/Al ratio as a chemical criterion and its critical value used in critical load calculations for forests, is insufficient.

Conclusion

Thus the study results comparable with the results of other authors indicate that Al can be toxic to plants but the degree of Al toxicity depends of plant species and especially its growth conditions.

The results showed that the yield of one-year-old pine growing in soil-water cultures was not related to the Ca/Al ratio in soil or soil solution or plants. Ca/Al ratio depends on the soil properties and was the lowest for Bv horizon.

In the sand-culture experiment the addition of 2 and 10 mg Al kg^{-1} did not affect biomass production, but a decrease of Ca/Al ratio in pine was observed. Increase of Al doses from 20 to 200 mg kg^{-1} decreased the yield of two-years-old pine and decreased Ca/Al ratio in plants. In that experiment a strict relation was found between the yields and Ca/Al ratios in needles, shoots and roots.

The toxicity indices given as Ca/Al value in the growth environment and in plants calculated for a given species under definite growth conditions are far from being universal, which testifies to our results and the results of other authors.

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