# Effects of Simultaneous Supply of Silicon and Boron on Plant Growth and on Herbicide Toxicity

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An optimal supply of biodisposable silicon and boron shows a synergistic stimulation effect on plant growth. Under these optimal growth conditions herbicides may become particularly toxic for cultural plants. Plants inhibited by herbicides show distinctly higher boron contents than that of untreated samples.

### Introduction

During our investigations on the effects of boron and silicon on plant virus infections the consideration of important interactions produced simultaneously by herbicides and both trace elements on plant became apparent [1].

It has become well known from extensive studies conducted under open air and greenhouse conditions that in order to optimize plant culture conditions, optimal mineral nutrition [2–4] combined with suitable herbicides [5, 6] is required.

Although the kinetics and the inhibition mechanism of many pesticides have been exhaustively studied, very little is known about the simultaneous influence of micronutrient concentrations in the culture medium on the plant growth and the efficiency of specific herbicides. Individual effects of both elements, boron [7–25] and silicon [25–31] in plant growth and cell physiology have been well studied. A large number of common vegetal species and cell cultures has been used. This paper deals with the simultaneous influence of silicon and boron on the growth of spinach hydrocultures and the action of the usual herbicides, viz., Stomp, Dicuran and Tribunil at variable Si/B supplies.

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### **Materials and Methods**

Spinach (*Spinacea oleracea*) of the variety "Matador" was used. The experiments were performed in 4 l hydrocultural synthetic pots containing the nutrient solutions according to Hoagland (Hoagland A + B, pH = 5.8) but lacking boron [32].

To the nutrient solutions, Si and B were added to final concentrations of 0/0, 25/0.3, and 50/0.6 (Si/B: ppm/ppm). Silicon was added as water glass and boron as borax. The stock solutions of Si and B were calibrated to pH = 5.5 with concentrated sulfuric acid avoiding any suspension or gel. The stock solutions of silicon were prepared immediately before the experiments.

The hydrocultures were automatically ventilated twice a day and additionally illuminated during 8 h per day by plant lamps of 600 Watts installed on a distance of 1.5 m. The water content was maintained constant by filling of with distilled water.

The spinach seedlings were put into each hydroculture on November 7 (1986) and the herbicides were added on November 21. The concentration of herbicides in the hydroculture solution was 10<sup>-6</sup> mol/l in all cases.

Our first experiments have been undertaken to study physiological relations and to measure appropriate analytical parameters. The major part of the plant material was harvested during growth period and used for chloroplast activity determinations.



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This work has been digitalized and published in 2013 by Verlag Zeitschrift für Naturforschung in cooperation with the Max Planck Society for the Advancement of Science under a Creative Commons Attribution-NoDerivs 3.0 Germany License. Another part was taken away for boron analysis and for isotope experiments. The observed dramatic effects of boron and silicon on growth stimulation and inhibition were unexpected. For these reasons, only a part of the plant material (and not the total final dry mass as indicated usually) was available at the end of the first experimental series (1986/87) for the biomass evaluation. Plant growth was also continuously followed and documented by photographs (Fig. 1a and 1b).

A new experimental series has then been started in November 1987, also under short day growing conditions. This time, the total vegetal dry matter has been determined in the end of the experiments, as done conventionally. Estimated green matter ratios from the first experimental series accord to the quantitative dry matter ratios in the second series. However, several hydrocultural pots have shown mycotic infections and produced incoherent results which could not confirm some of our first results. Fig. 2a and 2b refer to the mean biomass ratios from both experimental series.

Seedlings of spinach were brought in the state of germinated cotyledones into the hydroculture solutions already described (Nov. 1986 and 1987). All plants were cultivated in a greenhouse. Three series

of hydrocultures were grown. The first one contained neither silicon nor boron, the second one (medium concentration) 25 ppm Si and 0.3 ppm B and the third one (high concentration) 50 ppm Si and 0.6 ppm B. In this range, the last ratio of Si:B was found to be particularly favourable for spinach seedling growth. Two weeks later the specific herbicides were added to the corresponding hydrocultures leading to a final molar concentrations in the hydroculture of  $10^{-6}$  M.

Unfertilized white peat (TKS O) was filled into 5 l Mitscherlich pots (1.3 kg dry peat/pot) and brought to pH 5.5 with calcium carbonate. The basic nutrients were added, as well as one of the 3 different herbicide solutions (final concentration  $10^{-4}$  M), and Si and B solutions (final concentrations 0/0, 575/0.5, and 1150 ppm/1.0 ppm). The pots were left at room temperature for 2 weeks to reach the equilibrium. Thereafter,  $15 \times 3$  seeds were planted into each pot and watered regularly. Illumination conditions were identical as described for hydrocultures.

For the present study approximately 2,000 plants were used.

The chloroplast activity of the different samples was measured using the Hill reagent  $K_3$  [Fe(CN)<sub>6</sub>] [33].



Fig. 1a. Spinach hydrocultures photographed on December 1986 (shortly before cropping). The silicon and boron concentrations applied were 25 ppm and 0.3 ppm, respectively. The used symbols signify: O = no herbicide, S = Stomp, D = Dicuran, T = Tribunil. Each attempt was run twice.

D

S

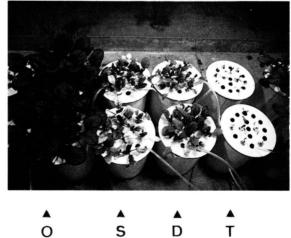


Fig. 1b: Spinach hydrocultures photographed on December 1986 (shortly before cropping). The silicon and boron concentrations applied were 50 ppm and 0.6 ppm, respectively (symbols: see Fig. 1a). Each attempt was run twice.

## Source of supply

Stomp (= N-(3-pentyl)-3,4-dimethyl-2,6-dinitro-aniline: Cyanamid GmbH), Dicuran (= 3-(3-chloro-4-methylphenyl)-1,1-dimethylurea: Ciba Geigy GmbH), Tribunil (= 1-benzothiazole-2-yl-1,3-dimethylurea: Bayer AG), water glass (Merck, DAB 6, according to the information of the producer the content of Si amounted to 27% SiO<sub>2</sub>), and borax (Sigma). Na<sub>2</sub>SO<sub>4</sub>: Merck p.a.).

## Boron analysis of leaves dry matter

For all plant material, the boron content was determined. 100 to 500 mg of dry matter was treated by pressure ashing method with 1–3 ml conc. HNO<sub>3</sub>, according to Schramel *et al.* [34]. The analyses were performed by inductively coupled plasma emissions spectroscopy (ICP) using the boron emission frequency of 49,68 nm [35]. The spectrometer was a JY 38<sup>+</sup> from Instruments SA (France).

## **Preliminary Results and Discussion**

The most remarkable results obtained from hydrocultures are documented in Fig. 1a and 1b. The biomass determinations after 28 and 42 days respectively are shown in the corresponding Fig. 2a and 2b. In the absence of both silicon and boron no growth of the seedlings took place under all conditions of hydroculture.

The largest amount of biomass was observed at high silicon and boron concentrations (50 ppm/0.6 ppm) without addition of any herbicide (:= 100%). Under these conditions, the biomass is approximately double of that found in medium Si/B concentrations and evidently more than the sum of the individual effects resulting separately from each of both trace elements.

We have also examined the contribution of sodium (introduced in our experimental system by the water glass supply) on spinach biomass production. Spinach is considered to be a sodiophilic plant. The following water cultures for silicon-free control experiments with sodium (Na<sub>2</sub>SO<sub>4</sub>) were used: i) 0.3 ppm B with 10 ppm Na<sup>+</sup> ("medium") and ii) 0.6 ppm B with 20 ppm Na<sup>+</sup> ("high"). No significant concentrations effects between the quite similiar hydroculture systems could be observed.

This is in agreement with literature describing salt stress effects on spinach growth over a large concentration range. At least in higher sodium concentrations spinach biomass was decreased [36, 37]. Ab-

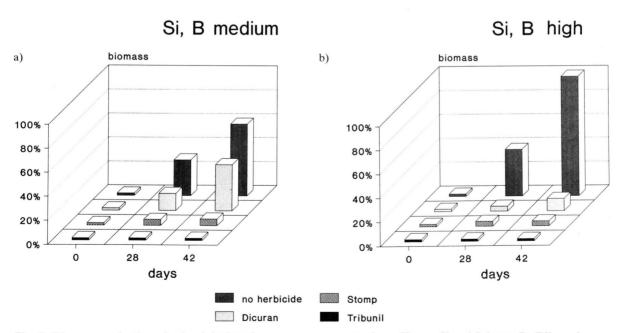


Fig. 2. Biomass production of spinach hydrocultures as a function of simultaneous silicon and boron supply. a) Medium concentrations: 25 ppm Si and 0.3 ppm B; b) high

concentrations: 50 ppm Si and 0.6 ppm B. Effect of some common herbicides: free of herbicides, Dicuran, Stomp and Tribunil. The highest biomass yield is set to 100%.

sence of effects [38] or opposite effects [39] are also reported at high supply of potassium.

Using our experimental scheme for other organisms we have also observed synergistic growth stimulation by boron and silicon supply for *Nicotiana tabacum* and *Scenedesmus subspicatus*.

Application of herbicides generally decreases biomass production more or less as a function of the type herbicide, of the plant and the environmental conditions [5, 6]. This has also been reported for spinach [40–46].

Synergistic phytotoxic effects of herbicides and boron have been investigated in field experiments. Reported results are inconsistent [19, 47–53].

In our case the addition of Dicuran (D), Stomp (S) and Tribunil (T), as typical examples of agricultural herbicides, and increasing concentrations of boron and silicon, lead to a distinct inhibitory effect. Moreover, the effects of the three herbicides are rather different. At medium concentrations of boron and silicon (Fig. 2a) Dicuran does not considerably reduce the growth of spinach whereas Stomp and Tribunil break down the metabolic activity of the plants (5% and 2% biomass, resp.).

At "optimal" concentrations of both trace elements (Fig. 2b) the biomass production of all herbicide treated plants is dramatically cut down. These results seem to be the first example of a quasi total plant growth inhibition at conditions which normally can be considered to be optimal.

The chloroplast Hill activity seems not to be affected if silicon and boron concentrations change from medium to higher values. In the case of medium contents of boron and silicon chloroplasts from herbicide treated plants show until to 30% less Hill activity compared to herbicide free samples.

Results of the boron content determinations in the plant material are given in Fig. 3. In dry matter of leaves from plants grown in the absence of herbicides an upper limit of boron concentration (15 ppm) in this case seems to be established. As we have found in other experiments using higher concentrations of boron, silicon seems to prevent the establishment of high boron concentrations in the leaves and also eliminate the toxic effects of excess boron in hydroculture and in soil.

The boron contents of spinach plants inhibited by simultaneous supply of herbicides and "optimal" boron and silicon concentrations are distinctly higher than herbicide free control plants (Fig. 3). Analo-

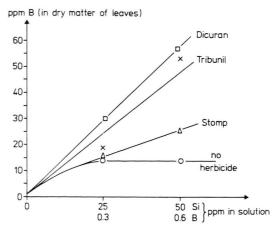


Fig. 3. Boron content in dry matter of spinach leaves as a function of simultaneous boron/silicon supply and the action of herbicides.

gous phenomenon has been reported for the boron content of cabbages treated by the herbicide Semeron [54]. In every case, the phenomenon of the combined boron-herbicide toxicity is strongly related to the simultaneous silicon supply.

Peat cultures are more comparable to convenient agricultural conditions than hydrocultures (because of adsorption and buffer capacity of soil). The effects observed show the same tendencies obtained in water culture but considerably less pronounced. As shown in hydroculture, there is a distinct difference between the inhibitor effects of Dicuran and the both other herbicides. The action of Dicuran on spinach growth is weaker and less dependant on boron/silicon supply.

In peat cultures all agents were added before sowing. Effects on germination process can be observed: Dicuran does not affect the germination. Stomp and Tribunil inhibit the germination during the first ten days at all boron/silicon concentrations chosen.

After this period, Stomp only allows the growth of very small and deformed seedlings whereas Tribunil at  $10^{-4}$  mol/l totally inhibits the germination.

### **Conclusions**

Biodisposable silicon and boron applied together can highly increase the biomass yields in spinach hydroculture. Under such conditions of accelerated biomass production, the plants can become extremely sensitive to herbicide application. Short day conditions and probably other stress parameters enhance the observed phenomenon.

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