

BUCKWHEAT STOMATAL TRAITS UNDER ALUMINIUM TOXICITY

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Abstract. Aluminium influence on some stomatal parameters of common buckwheat (*Fagopyrum esculentum* Moench.) was studied. Significant changes in stomatal density, stomatal index and stomatal shape coefficient under aluminium treatment were revealed. Stomatal closure and no difference in total stomatal potential conductance index of treatment plants were suggested as aluminium resistance characteristics.

Key words: *Fagopyrum esculentum*, common buckwheat, stomata, aluminium toxicity, aluminium resistance

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Introduction

Aluminium (Al) toxicity is the most significant factor reducing crop production worldwide. Al is highly active in acid soil (pH < 5.0) and toxic for plant growth (KOCHIAN *et al.* 2004). The first symptoms of aluminium stress are disruptions of growth parameters: inhibition of root and shoot elongation, decrease in total plant biomass and diminution of leaf area. A change in morphological parameters is the response of plant to aluminium toxicity. Rate of this change can be used for characterization plant aluminum resistance (SILVA *et al.* 2012). Meanwhile the changes in the leaf anatomy as induced by aluminium are model in many plants due to the same defense mechanisms. Stomatal parameters such as stomatal density, size, potential conductance and closure also can be used as stressful condition signs and indicators of aluminium resistance (ÖZYİĞİT & AKINCI 2009).

Material and methods

Seeds of common buckwheat (*Fagopyrum esculentum* Moench.) were germinated in the dark at 25°C in Petri dishes with deionized water. After 2 days seedlings were transferred in pots with sterilize sand and ½ Hoagland solution (pH 6.5) in trays. On 7 day the

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aluminum (50 µM) was added to ½ Hoagland's solution that did not contain phosphorus with full strength micronutrients. Each day the solutions with Al were adjusted to a pH 4.5. The experiment was conducted in a growth room with controlled temperature (25±2°C), and a photoperiod of 16 hours.

Ten days after treatment with Al, stomatal parameters were determined on adaxial (top) and abaxial (lower) leaf surface of common buckwheat using imprints made with nail enamel. Imprints were observed through Bresser LCD Microscope and photographed. Anatomic investigation of stomata included: stomatal density per mm² (SD), stomatal index (SI) (ROYER 2001), stomatal length (SL), stomatal width (SW) (MAHERALI *et al.* 2002), stomatal surface (SS), stomatal shape coefficient (SSC) and potential conductance index (PCI) (WANG *et al.* 2012).

For statistical analysis the data were subjected to paired-sample *T*-tests, using Microsoft Excel 2007, with 99% ($P \leq 0.01$) and 95% ($P \leq 0.05$) least significance of differences between means.

Results and discussion

Stomatal movement and functional status of the guard cells are one of the important regulation factors in the connections between plant and environmental state. Studied stomatal

Table 1. Changes of stomatal parameters under aluminium treatment.

Parameters	control		aluminium (50 μM) treatment	
	adaxial surface	abaxial surface	adaxial surface	abaxial surface
stomatal density (n/mm ²)	55	68	50*	47*
stomatal index	16.4	18.17	15.2**	15.7*
stomatal length (μm)	33.64	30.28	29.85*	40.57*
stomatal width (μm)	23.28	23.57	19.85*	22.85**
stomatal surface (μm^2)	614.76	560.25	465.29	727.71
stomatal shape coefficient	69.20	77.84	66.50	56.32
potential conductance index	6.22	6.23	4.45	7.73

Significant at: *p=0.01, **p=0.05

parameters are very sensitive to changes of growth conditions – temperature (BEERLING & CHALONER 1993), CO₂ concentration (ROYER 2001), water stress and soil salinity (BUCKLEY 2005), heavy metal toxicity (ABDUSSALAM *et al.* 2013).

Buckwheat is highly resistance to aluminum stress and is known to be a bioaccumulator of aluminum (SHEN *et al.* 2006). It is well known mechanisms of buckwheat external and internal aluminum detoxifying by oxalic acid (KLUG *et al.* 2011). But aluminum effect on the reaction of stomatal parameters is not yet completely understood.

Aluminum effects on stomatal parameters of *F. esculentum* leaves are presented in Tab. 1. It was observed that stomatal density and stomatal index were significantly decreased under aluminum treatment.

The greatest influence of 50 μM Al was revealed at abaxial leaf surface on SD, SI and SSC parameters which were 69%, 86% and 72% of control values. The maximum inhibition of stomatal surface was observed at adaxial side – 83% of control value. Reduction of stomatal density per mm² could be a consequence of aluminum effect on protodermal sister cell division into guard cells. The similar effect of cadmium was detected in burley plants (KAZNINA *et al.* 2011).

Changes in stomatal shape coefficient under aluminium treatment directly connected with stomatal closure (Fig. 1 A-C). Stomatal movement under aluminium stress is the signal of transpiration inhibition. Thus, it should

be noted that stomatal length and stomatal surface at abaxial leaf side of treatment plants were increased on 33% and 29% respectively. Such changes of these parameters allowed removing the total (two-sided) stomatal potential conductance to the control value (see Tab. 1). Received data can be an assertion of the buckwheat resistance to aluminium stress that confirmed by activation of adaptive mechanisms that control functional status of the stomatal apparatus.

References

- ABDUSSALAM A.K., RATHEESH CHANDRA P., KOORIMANNIL H., SALIM N. 2013. Response and bioaccumulation potential of *Boerhavia diffusa* L. towards different heavy metals. *J. Stress Phys. Biochem.* 9 (3): 23–36.
- BEERLING, D., CHALONER W. 1993. The impact of atmospheric CO₂ and temperature change on stomatal density: observations from *Quercus robur* lammas leaves. *Ann. Bot.* 71: 231–235.
- BUCKLEY T. 2005. The control of stomata by water balance. *New Phytol.* 168: 275–292.
- KAZNINA N., TITOV A., LAIDINEN G., BATOVA J. 2011. Effect of cadmium on water relations in barley plants. *Transaction of Karelian research centre of Russian Academy of Science* 3: 57–61. (in Russian).
- KLUG B., SPECHT A., HORST W. 2011. Aluminium localization in root tips of the aluminium-accumulating plant species buckwheat (*Fagopyrum esculentum* Moench). *J. Exp. Bot.* 62 (15): 5453–5462.
- KOCHIAN L., HOEKENGA O., PINEROS M. 2004. How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorous efficiency. *Ann. Rev. Plant Biol.* 55: 459–493.

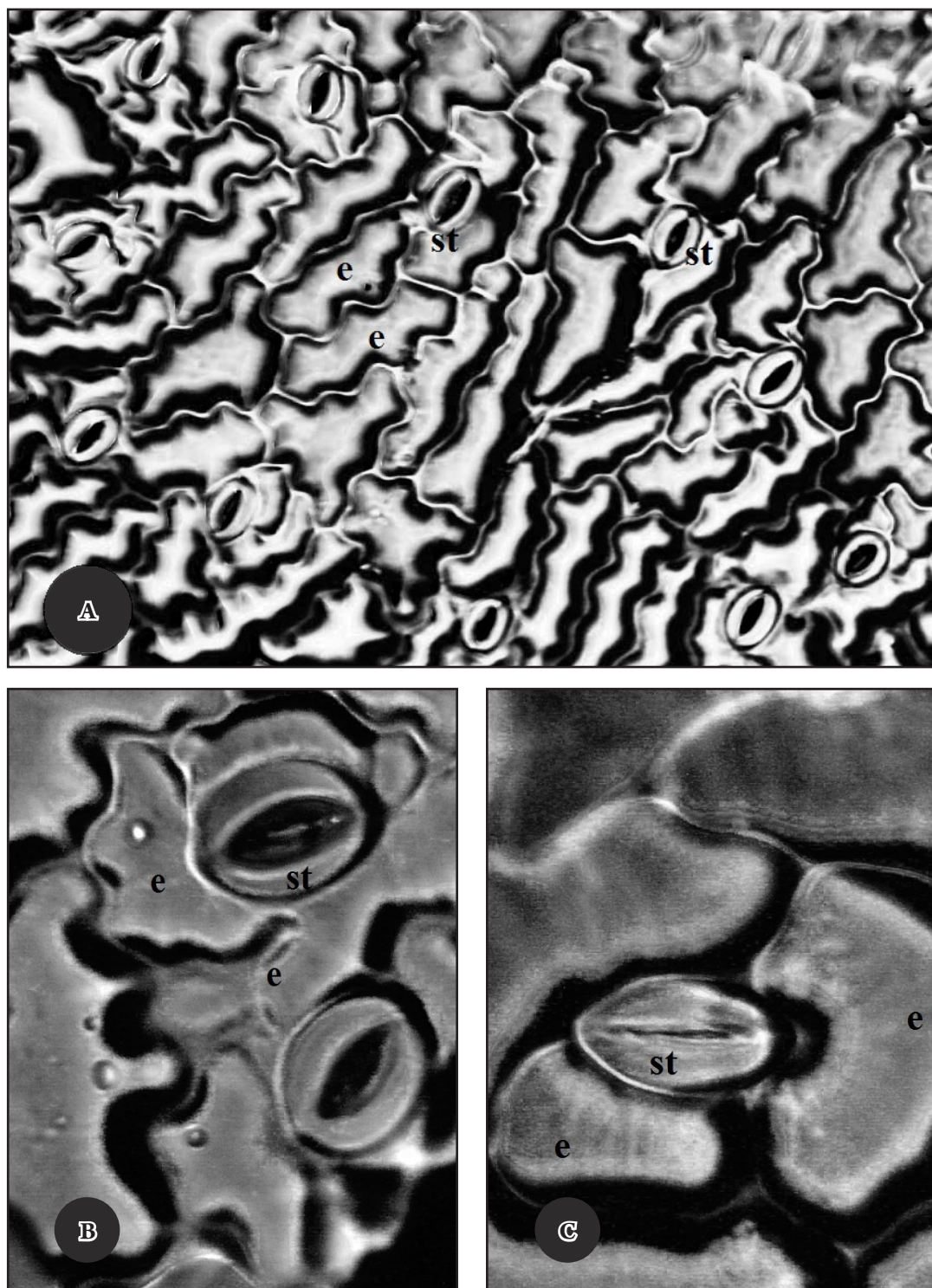


Fig. 1. Stomatal apparatus of buckwheat (*Fagopyrum esculentum*) leaves: **A** – stomatal density at abaxial surface of control plants; **B** – opened stomata of control plants; **C** – stomatal closure under aluminium treatment. **e** – epidermal cell; **st** – stoma.

- MAHERALI H, REID C., POLLEY H., JOHNSON H., JACKSON R. 2002.** Stomatal acclimation over a subambient to elevated CO₂ gradient in a C₃/C₄ grassland. *Plant Cell Environ.* **25**: 557–566.
- ÖZYİĞİT I., AKINCI S. 2009.** Effects of some stress factor (aluminium, cadmium and drought) on stomata of roman nettle (*Urtica pilulifera* L.). *Not. Bot. Hort. Agrobot. Cl.* **37**: 108–115.
- ROYER D. 2001.** Stomatal density and stomatal index as indicators of paleoatmospheric CO₂ concentration. *Rev. Palaeobot. Palynol.* **114**: 1–28.
- SHEN R.F., CHEN R.F., MA J.F. 2006.** Buckwheat accumulates aluminum in leaves but not in seeds. *Plant Soil* **284 (1-2)**: 265–271.
- SILVA S., PINTO G., DIAS M., CORREIA C., MOUTINHO-PEREIRA J., PINTO-CARNIDE O. SANTOS C. 2012.** Aluminium long-term stress differently affects photosynthesis in rye genotypes. *Plant Physiol. Biochem.* **54**: 105–112.
- WANG H., SHI H., YANG R., LIU J., YU Y. 2012.** Stomatal characteristics of greening plant species in response to different urban atmospheric environments in Xi'an China. *J. Food Agric. Env.* **10 (3-4)**: 1524–1529.