

Physiological characterization of salt-tolerant alfalfa (*Medicago sativa* L. and *Medicago sativa* spp. *falcata*)

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Increasing soil salinity is a global issue that threatens crop production. Alfalfa (*Medicago sativa* L.) is a major forage crop worldwide that is relatively sensitive to soil salinity. Improved cultivars with high production on saline soil will benefit many producers and land managers. Utilizing a greenhouse selection protocol, we developed several lines of alfalfa with the ability to survive high concentrations of salinity after three cycles of selection. This study reports the initial physiological characterization of two experimental lines, CS15-2 and BC11-1. Based on stem length and leaf number measurements, both demonstrated better growth under salt treatment compared to their parents. This improved growth is associated with greater accumulation of chlorophyll contents in the experimental lines measured by the chlorophyll content index. The experimental lines also showed an improved capability to maintain water status (relative water content). The two selection lines, however, showed differences in Na⁺ accumulation after salt treatment; the Na content increased in CS15-2 but did not change in BC11-1 as shown in their respective parents. The differences suggest that these two selection lines may have different mechanisms in regulating Na uptake and transport but the difference in Na content in the shoots may not be the key to determining their salt tolerance. Data indicated that CS25-2 significantly accumulated other inorganic ions such as Ca²⁺, K⁺, and PO₄³⁻ while its parents did not accumulate or even decrease the accumulation of these ions, suggesting CS25-2 may be better at maintaining a balance of different ions. BC11-1 did not show a significant difference from its parents in change in these ions during salt treatment. These results provide a physiological basis to support the improved salt tolerance in these selection lines. The results also suggest the two selection lines may have adapted different mechanisms in salt tolerance.