

Adaptation to Abiotic Stress Factors

[Mechanisms of Adaptation of *Brachiaria* Cultivars to Aluminum-toxic and Low-fertility Acid Soils] [Screening Method to Identify Aluminum Resistant *Brachiaria* Hybrids]

I. Mechanisms of Adaptation of *Brachiaria* Cultivars to Aluminum-toxic and Low-fertility Acid Soils

At CIAT there is an ongoing program to genetically improve *Brachiaria* cultivars, which seeks to combine favorable traits such as adaptation to acid soils (*B. decumbens*), spittlebug resistance (*B. brizantha*) and nutritional quality (*B. ruziziensis*) within new apomictic cultivars. Such cultivars would provide farmers with an environmentally attractive low-cost alternative to chemical control of the pest for *B. decumbens* or application of substantial amounts of fertilizer for *B. brizantha*, neither of which is desirable or feasible for low-input livestock production systems.

Progress in the development of superior *Brachiaria* cultivars is strongly dependent on the availability of rapid and efficient screening procedures to genetically recombine the desirable traits. Compared to screening methods available for spittlebug resistance, apomictic reproduction and nutritional quality, the methods for evaluating acid soil adaptation are considerably less advanced, thus providing a substantial obstacle to the progress of the genetic improvement program. The main reason for this is that there are several growth-limiting factors in infertile acid soils, the most important being aluminum (Al) toxicity and the deficiency of nutrients such as phosphorus (P), nitrogen (N), calcium (Ca) and magnesium (Mg). Until recently, it was not known, which of these factors determine the interspecific differences in acid soil adaptation observed under field conditions. Even more problematic was the fact that these differences are observed only after 2 to 3 years of establishment of pastures.

The classical method of screening for adaptation to infertile soils is based on forage yield in the field, which is very expensive and time-consuming. Consequently, indirect methods, based on physiological or biochemical responses were needed. For their development, a system to simulate acid soil stress under well-defined conditions on a short-term time scale was required. Therefore, we designed a low ionic-strength nutrient solution that simulated the nutrient-deficient and Al-toxic conditions in soil solutions from acid soils collected from the

Colombian savanna region. Interspecific differences in growth observed when seedlings were cultivated in this solution for 2 to 3 weeks were similar to growth differences in the field observed 1 to 3 years after pasture establishment.

Based on these results, we used this "acid-soil" treatment as a point of departure to develop several lines of research. The main aim of this research was to dissect the "acid soil syndrome" into individual components that were assumed to be most relevant for *Brachiaria* cultivars: Toxicity of Al and deficiency of P and N. For each of these factors, several hypotheses concerning physiological and biochemical traits possibly contributing to the excellent adaptation of *B. decumbens* to infertile acid soils were developed and tested. The aim was to assemble a set of traits that could be used for the design of a screening procedure for acid soil adaptation. We implemented a broad array of new methods required, and when necessary we sought collaboration of specialized laboratories outside of CIAT. The progress made thus far is summarized below.

Mechanisms of Al Resistance

- Although there is a significant interspecific variation in Al resistance among *Brachiaria* species, their level of Al tolerance is markedly superior than that of all so far investigated Al-resistant crop varieties;
- *B. decumbens* exhibits an outstanding level of Al resistance;
- Surprisingly, this high level of Al resistance is achieved in virtual absence of organic acid exudation by root tips, contrary to the current view of Al resistance mechanisms;
- *B. decumbens*, but not the non-adapted cultivar *B. ruziziensis*, seems to detoxify Al efficiently within tissues by chelation with different organic acids: malic acid in root tips, citric acid in mature parts of the root system and oxalic acid in shoots;
- P transport into root tips is inhibited by Al in *B. ruziziensis* but not in *B. decumbens*;
- All cultivars strongly increase the rhizosphere pH around the elongation zone of root tips (the most Al-susceptible part of the root), which is expected to sharply decrease Al solubility;
- Root cell wall cation exchange properties are not related to the interspecific differences in Al resistance.

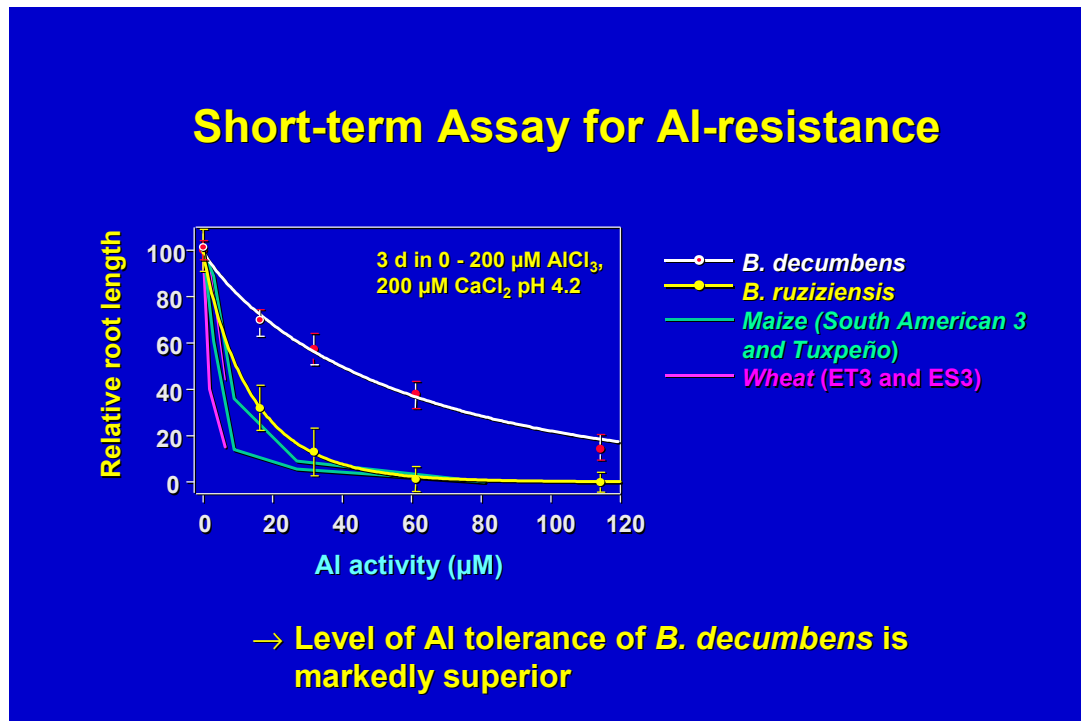


Figure 1. Level of Al resistance of *Brachiaria decumbens* compared with *B. ruziziensis* and two cereal crops (maize and wheat). This assay involves use of seed to generate seedlings and the seedlings were exposed to Al for 3 days.

Traits Contributing to Adaptation to P-deficiency

- The root system of *B. decumbens* contains considerably finer and longer roots than that of the other cultivars, suggesting that *B. decumbens* can take up more P per unit leaf area to support photosynthetic activity under P-deficiency;
- The high root-surface acid phosphatase activity is likely to enhance P uptake from organic sources in all *Brachiaria* cultivars;
- The rate of internal P turnover appears to be the highest in *B. decumbens* as suggested by the low tissue P_i concentration and the high internal acid phosphatase activity.

Adaptation to Low N Supply - we found that:

- *B. decumbens* has a considerably lower external N requirement than the other cultivars, which was concluded to be the result of a markedly superior NO_3^- uptake efficiency,

- Roots of all *Brachiaria* cultivars accumulate large quantities of two aromatic secondary metabolites under N and P deprivation, which were identified as 1,3-di-feruloylquinic acid and 1-feruloyl-3-coumaroylquinic acid, two hitherto unknown phenylpropanoid derivatives,
- Accumulation of these compounds is positively correlated among *Brachiaria* cultivars with sensitivity to acid soil stress.

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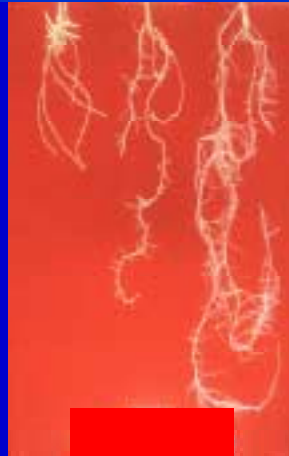
II. Screening Method to Identify Aluminum Resistant *Brachiaria* Hybrids

We adapted the seedling bioassay to vegetative cuttings. We demonstrated that measuring root elongation, using stem cuttings in a solution containing 200 μM CaCl_2 , 200 μM AlCl_3 (pH 4.2), is a simple, but powerful method to discriminate between *B. decumbens* and the other two cultivars. Al toxicity markedly reduced root elongation of *B. brizantha*, while it had little effect in *B. decumbens*, confirming the high level of Al resistance of the latter. *B. ruziziensis* is significantly less Al-resistant than *B. decumbens*. However, this difference was not noticeable in the assay, because roots of *B. ruziziensis* hardly elongated in the solution lacking Al. Only roots of *B. decumbens* continued to elongate under simultaneous Al stress and nutrient deficiency.

Screening for aluminum resistance in *Brachiaria* (Vegetative cuttings)

21 days with
200 μ M Al
in solution

B.ruziziensis *B.brizantha* *B.decumbens*



We are using this screening method to identify most promising *Brachiaria* hybrids that combine Al resistance with spittlebug resistance. We have identified 2 sexual hybrids (SX 2349 and SX 497) with greater level of Al resistance than that of the sexual parent, BRUZ/44-02. Among the apomictic hybrids, CIAT 36061 showed greater level of Al resistance but its level of Al resistance is significantly lower than that of *B. decumbens*. A new cycle of hybrids are being evaluated to identify apomictic hybrids that combine high level of Al resistance with spittlebug resistance.

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