

# English Version

## Abstract

The use of forages with aluminum tolerance is the best alternative to the wide areas of acid soils with high aluminum concentrations in Brazil. So, the goals of this work were: to evaluate the aluminum level in the nutrient solution and the most important characteristics for elephant grass genotypes discrimination; to verify the existence of genetic variability between accesses of elephant grass of BAG Embrapa Gado de Leite for aluminum tolerance, and; to study the genotypes and aluminum levels interaction. In a greenhouse experiment 26 elephant grass genotypes and four aluminum levels (0, 15, 30 e 45 ppm) were tested, using a randomized design, with three replications and only one plant per vase. Data of dry matter of aerial part (DMAP), root dry matter (RDM) height of aerial part (HAP) and root length (RL) were collected. Using the methodologies of Eberhardt and Russell and Annicchiarico, analysis of adaptability and stability were done using the averages of the genotypes on the different levels of aluminum. There was variability between elephant grass genotypes for tolerance to the different aluminum levels, regarding the characteristics DMAP, RDM, HAP and RL. The level of 15 ppm of aluminum was considered the most appropriated to identify tolerant and sensible genotypes to aluminum toxicity. DMAP and HAP were the characteristics indicated for future evaluations of elephant grass for aluminum tolerance using nutrient solution. Taiwan A 121 and Australiano genotypes presented adaptability and stability in relation to the two employed methodologies.

**Key word:** *Pennisetum purpureum*; forage; genetic breeding

## Aluminum toxicity tolerance in elephant grass<sup>1</sup>

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## Introduction

With the increase of the competitiveness of the productive sector, dairy quest, by all forms, for production systems which provide higher yield and fewer costs. Among the strategies adopted with this goal is the improvement of the herb and fodder quality provided to the animals, so that they express all its productive potential.

The high potential yield of elephant Grass fodder associated to other fodder characteristics appropriated, i.e. high quality, palatability, vigor and persistency, has stimulated not only the harvest of this species but also its breeding aiming to develop cultivars to use to graze and grass.

Elephant Grass presents high potential to increase in production of milk (DERESZ, 1990) and meat (SETELICH et al., 1998). It is widespread throughout Brazil, and it can be cultivated in divergent environmental conditions, although there

are few improved cultivars available, especially for the use under rotational grazing.

In Brazil most of the soils destined to the vegetal production have low fertility and problems with acidity and toxicity by aluminum, which are factors responsible for the low yield of most of the crops. On the specific case of grazing, which normally occupies marginal areas, these problems are more serious. The high aluminum concentration on the acid soils assumes, therefore, an important part on national agriculture and livestock, affecting directly the physiologic and metabolic processes of most of the cultivated species.

Among the alternatives considered viable to overcome these difficulties, the obtaining of improved cultivars, tolerant to toxic aluminum, has been considered the most promising, and it constitutes one of the most important demands of the milk producers of all country. It is intense the demand for fodder varieties adapted to the different ecosystems.

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To most of the cultivated species it is verified wide variability on the behavior under stress conditions caused by aluminum. To some crops, there are reports on the literature of the possibility of the selection of materials more adapted to these conditions, performed in nutrient solution, as, for instance, to corn (CAMBRAIA and CAMBRAIA, 1995; PINTRO et al., 1996; MAZZOCATO et al., 2002), soy bean (MENOSSO et al., 2000), rice (FERREIRA et al., 1998), wheat (CAMARGO et al., 1998 and CAMARGO and FERREIRA, 1999) and bean (SANTOS et al., 1997). To elephant grass, it was not found reports about its behavior concerning aluminum toxicity.

Considering what was shown, the objectives of the present work were: a) to evaluate the level of aluminum of the nutrient solution and the most important characteristic to the discrimination of genotypes of elephant grass; b) to verify the existence of genetic variability between some Elephant Grass accessions on the BAG on the Embrapa Gado de Leite to aluminum tolerance and; c) to study the interaction genotypes per aluminum levels.

## Material and Methods

The experiments were conducted on Embrapa Gado de Leite, located on the municipality of Juiz de Fora (MG), on the Longitude 43° 15' 44" West, Latitude 21° 35' 04" South and 426 m of altitude.

Twenty six elephant grass genotypes from the Banco Ativo de Germoplasma of the Embrapa Gado de Leite were used (Table 1). From each of these accessions seedlings were obtained, in greenhouse, through rooting of shoots in sandboxes. After the

rooting period (20 days), the seedlings were selected according to the uniformity of size of the shoots and roots, and transplanted into plastic pots with a volume of two liters. It was used, as substrate, the Clark nutrient solution (1975), modified. In this solution, the only variable element was the aluminum, used in four different levels: 0, 15, 30 and 45 ppm.

The experiment was installed in a greenhouse on the completely randomized design, with three repetitions and pots of a vase with a plant. In intervals of three days it was performed a rotation on the vase position in the stands to assure homogeneity on the environment conditions.

Twenty five days after the transplanting, plants were collected and divided in roots and shoots. Data referent to the dry mater of the shoots (PSPA), dry matter of the roots (PSR), height of the shoot (APA) and root length (CR) was recorded. For each characteristic it was performed an analysis of variance, considering the completely randomized design in factorial treatment. With the averages of the genotypes on different aluminum levels it was performed an analysis of the adaptability and stability using the methodologies described by Eberhart and Russell (1966), according to Cruz and Regazzi (1997), and Annicchiarico (1992).

To identify the best aluminum level of the nutrient solution to the Elephant grass genotype discrimination it was performed variance analysis (DIC) to each level used. It was based on the criterion proposed by Ferreira et al. (1998), i.e.: 1) maximum value of the statistic F; 2) maximum value of genotype determination coefficient ( $R^2$ ) and 3) maximum value of the relation between the variation coefficient of genetics ( $CV_g$ ) and experimental ( $CV_e$ ).

**Table 1.** Elephant Grass genotypes used to determine the level and the most important characteristic to genetic studies about the tolerance to aluminum toxicity.

1) Gigante de Pinda	10) Sem Pêlo	19) IJ 7136
2) Elefante Híbrido 534 A	11) Mott F1	20) Australiano
3) Mercker Pinda	12) Cuba 115	21) 13 AD
4) Mercker Pinda México	13) Cuba 169	22) 10 AD
5) Mercker Comum Pinda	14) Cameroon	23) 12 AD
6) Taiwan A 121	15) Napierzinho	24) HV 290 23a x Elefante
7) Mole de Volta Grande x 239 DA	16) IJ 7125	25) 02 AD
8) Mineiro x 23 A	17) IJ 7126	26) 08 AD
9) Elefante Cachoeiro de Itapemirim	18) IJ 7127	

## Results and discussion

Initially it must be mentioned the good experimental precision, evaluated by the estimate variance coefficient (CV) of the statistic analysis to the different characteristics evaluated. It was observed an oscillation on the magnitude of the estimates of 7.5% to the height of shoots, and 18.1% to the dry matter of roots. In accordance with Pimentel Gomes (1990), these estimates are considered low to medium, showing the experiment accuracy, which reinforces the reliability of the results obtained.

Significant differences between genotypes and aluminum level were observed to all the characteristics evaluated, which indicate the existence of variability between the elephant grass accessions to toxic aluminum tolerance and between the aluminum levels tested. The interaction between genotypes and aluminum levels was also significant in all cases, showing the inconsistency of the genotype behavior on different aluminum levels for PSPA, PSR, APA and CR. These results justify the study about stability and adaptability of elephant grass genotypes, on the interaction detailing.

The results of the average performance of the elephant grass clones to PSPA, PSR, CPA and CR in each aluminum level are shown on Table 2. It was verified, for all the statistics, a decrease on the averages obtained with the increase on the aluminum concentration. In spite of the interaction between genotypes and aluminum levels observed by the alteration on the genotype classification on different levels, it was noticed that some genotypes present performance relatively stable. It is the case, for instance, of the group 08 AD, which was always on the group statistically superior to PSPA on the different aluminum levels tested.

Considering the genotype average involving different aluminum genotypes, it was also observed large variations on the performance for all statistics. To PSPA, the genotype averages were divided in six distinct groups, by Scott and Knot test. The variation amplitude between averages was 3.1 g, superior to the overall experiment average (3.01 g). Genotypes 08 AD, AD, IJ 7126, Cameroon, IJ 7125, IJ 7136 and Napierzinho constituted the group with best average performance (Table 2)

Regarding to PSR, it was found the value

of amplitude variation of 0.6 g. The genotypes were divided in five distinct groups, and the clone IJ 7136 was the one that presented higher average weight of roots, with 1.1 g. To shoot height and CR it was identified six and four groups statistically different, by Scott-Knott test. Genotypes Cameroon and 02 AD were the only classified on the group with superior performance to both characteristics (Table 2)

The use of different aluminum levels in studies of tolerance or resistance to this element toxicity makes it difficult or unfeasible to evaluate a large number of genotypes, since it demands more resources and adequate infrastructure. In the identification of ideal environmental conditions for the performance of genetic studies it is considered as the most appropriated ones those that provide good experimental accuracy and allow larger expression of genetic variability (FERREIRA et al., 1998). Thus, it was searched, between the aluminum levels studied, the one with lower experimental variance coefficient and high estimates on the relation  $CV_e/CV_g$ ,  $R^2$  and statistic F.

With regard to the different Al levels studied, it was observed variation in each estimate (Table 3).  $CV_e$ , for instance, ranged from 12.6% to PSPA at level 15 ppm to 25.9% at level 45 ppm of aluminum. By contrast the coefficient of determination to the CR ranged from 0.65 at level 0, to 0.94 at level 45 ppm. Overall, the level 15 ppm of aluminum was considered the most appropriated to identify materials tolerant and sensitive to aluminum toxicity since it presented the highest values of statistics F, relation between coefficient of genetic variation and experimental, highest coefficient of genotype determination and lower coefficient of environmental variation.

It is noteworthy that in most of the levels tested the highest coefficients of determination were obtained to PSPA and APA. The only exception is referent to the level 45 ppm in which Cr presented, together with APA, the highest  $R^2$ . It was observed, however, that the  $R^2$  to PSPA was also high at this level. These results indicate that PSPA and APA provided better adjustments of data to the model. The association of this information with the facility and practicality of the evaluation and measurement of these two characteristics, it is suggested the use of only these two characteristics in future works of

**Table 2.** Averages of dry matter of shoots (PSPA), dry matter of roots (PSR), height of shoots (APA), and root length (CR) of elephant grass genotypes evaluated in nutrient solutions with different aluminum levels.

Genotype	PSPA (g)	PSR (g)	APA (cm)	CR (cm)
08 AD	4.6a	1.0b	67.1d	37.0b
10 AD	4.4a	0.9b	77.1b	43.5a
IJ 7126	4.3a	1.0b	63.5d	45.3a
Cameroon	4.2a	1.0b	79.5a	43.7a
IJ 7125	4.1a	1.0b	74.27c	46.1a
IJ 7136	4.0a	1.1a	73.6c	43.5a
Napierzinho	4.0a	0.8c	76.7b	35.4c
Taiwan A 121	3.7b	0.8c	76.98b	34.5c
Mineiro x 23 A	3.7b	0.6d	76.5b	41.3a
Australiano	3.5b	0.9c	83.4a	30.3d
02 AD	3.4b	0.8c	84.0a	43.3a
13 AD	3.1c	0.7d	75.3b	30.5d
Elefante Híbrido 534 A	3.0c	0.7d	80.6a	29.1d
Gigante de Pinda	2.9c	0.8c	82.0a	34.3c
Mott F1	2.6d	0.5e	70.5c	37.0b
Sem Pêlo	2.5d	0.7d	61.5d	39.4b
Mercker de Pinda	2.5d	0.5e	62.0d	32.8c
Mole de Volta Grande x 239 DA	2.3e	0.7d	64.6d	32.0c
Mercker Pinda México	2.2e	0.7d	65.3d	30.3d
Cuba 169	2.1e	0.7d	74.7b	29.6d
Cuba 115	2.0e	0.8c	71.9c	25.2d
12 AD	2.0e	0.5e	50.4f	27.8d
Elefante Cachoeiro de Itapemirim	1.9f	0.6e	56.3e	35.3c
Mercker Comum Pinda	1.8f	0.7d	64.1d	32.7c
IJ 7127	1.8f	0.4e	57.7e	32.1c
HV 290 23a x Elefante	1.5f	0.5e	53.48f	37.8b
Média Geral	3.01	0.75	70.11	35.75

*Averages followed by the same letter in the column do not differ by the Scott Knott test (1974) at 5% of probability*

evaluation of elephant grass tolerance to aluminum.

As it was previously mentioned, the interaction between genotypes and aluminum levels was significant, and enabled a most detailed study of the genotype behavior on different levels, when trying to identify adapted and stable materials. Since PSPA and APA were the characteristics recommended to future evaluations, the results of the adaptability and behavior stability will be discussed only in function of them.

According to the methodology described by Eberhart and Russel (1966), cited by Cruz and Regazzi (1997), cultivars of overall adaptability are those that present regression coefficient ( $\beta_{ii}$ ) equal to 1. Regression coefficient lower than 1 indicates

cultivars adapted to unfavorable environments and higher than 1 shows cultivar of stable behavior and responsive to environmental improve.

To shoot dry matter the genotypes Mercker de Pinda, Taiwan A 121, Sem Pêlo, IJ 7127, Australiano, 13 AD and 12 AD presented  $\beta_{ii}$  significantly higher than the unit (Table 4). It was observed that, among these materials, genotypes IJ 7127 and 12 AD presented lowest PSPA averages. Conceptually it is considered material of good adaptability that with good productive performance. With this in mind, although these two materials have presented regression coefficient superior than the unit, they cannot be considered of good adaptability in relation to the toxic aluminum. Moreover, the predictability

**Table 3.** Coefficient of experimental variation ( $CV_e$ ), coefficient of genotype determination ( $R^2$ ), coefficient of genetics variation ( $CV_g$ ), ratio of coefficient of genetics variation/coefficient of experimental variation ( $CV_g/CV_e$ ) and statistic  $F$  of the characteristics: dry matter of shoots (PSPA), dry matter of roots (PSR), height of shoots (APA), and root length (CR)

Level	Characteristic	$CV_e$	$R^2$	$CV_g$	$CV_g/CV_e$	F
0	PSPA	16.5	0.88	26.6	1.6	8.8
	PSR	16.3	0.84	21.9	1.3	6.4
	APA	5.4	0.88	8.7	1.6	8.7
	CR	14.0	0.65	11.0	0.7	2.8
15	PSPA	12.6	0.95	34.0	2.6	22.7
	PSR	13.3	0.92	27.8	2.0	13.9
	APA	5.8	0.92	12.2	2.1	14.2
	CR	16.7	0.69	14.6	0.8	3.2
30	PSPA	19.9	0.92	40.0	2.0	13.0
	PSR	20.5	0.90	36.9	1.8	10.6
	APA	8.8	0.92	17.6	1.9	12.9
	CR	15.3	0.85	21.7	1.4	7.0
45	PSPA	25.9	0.92	51.0	1.9	12.5
	PSR	28.1	0.90	49.2	1.7	10.1
	APA	11.2	0.94	26.6	2.3	17.8
	CR	19.2	0.94	45.2	2.3	17.5

of behavior of these two genotypes, estimated by the determination coefficient ( $R^2$ ), was lower than the others. In this aspect, the genotype Gigante de Pinda presented the behavior most unpredictable, with  $R^2$  of 22.5%, and it was considered little stable. This genotype also presented  $\beta_{ii}$  lower than 1 ( $P < 0,01$ ), showing good adaptability to unfavorable conditions, i.e., good PSPA production on elevated aluminum levels.

The results of the adaptability and stability evaluation of elephant grass genotypes using the accuracy index (ANNICCHIARICO, 1992), are presented on Table 4. It was observed that various materials presented accuracy estimates superior to 100%, indicating that its use is associated to low probability of failure. Genotype 08 AD presented lower risk ( $Wi = 153.1\%$ ), showing that its behavior was 53.1% superior to the environmental average, with 75% probability. Furthermore, it was verified that most of the genotypes with high PSPA production (high average) presented high accuracy level. Genotypes Taiwan A 121 and Australiano were the only ones that proved to be adapted and stable by the evaluation parameters of the two

methodologies.

The results obtained in this work show the large range observed to all the characteristics studies, confirming the existence of genetic variability between elephant grass accessions and the effects of aluminum levels on this species development. Moreover, it allows to foresee the possibility of success with the selection of elephant grass genotypes tolerant to toxic aluminum and with productive behavior appropriated and stable in different levels of this element. Although, since all the experiments were performed in Green house using nutrient solution to simulate the possible different environment conditions, it is necessary to confirm these results with the performance of field experiments. In this case, however, the number of genotypes tested may be smaller, and their choice can be based on this work. Once it is confirmed the efficiency of this methodology to identify genotypes tolerant to aluminum, the next step is an evaluation of all the elephant grass Active Germplasm Bank of Embrapa Gado de Leite, aiming to its characterization and later use to breeding of this fodder.

**Table 4.** Estimates of adaptability and stability parameters of elephant Grass genotypes, accuracy index (Wi), considering the dry matter of the shoots (PSPA) in four aluminum levels

Genotype	$\beta_0$	$\beta_{1i}$	R <sup>2</sup>	Wi
Gigante de Pinda	2.13	0.48**	22.52	61.99
Elefante Híbrido 534 A	3.02	1.36*	99.60	90.81
Mercker Pinda	2.48	1.50**	95.03	63.87
Mercker Pinda México	2.24	0.59**	99.76	75.14
Mercker Comum Pinda	1.80	0.31**	79.37	60.36
Taiwan A 121	3.74	1.54**	99.18	115.21
Mole de Volta Grande x 239 DA	2.31	0.29**	98.31	77.96
Mineiro x 23 A	3.66	0.78	92.50	122.79
Elefante Cachoeiro de Itapemirim	1.89	0.92	83.04	50.66
Sem Pêlo	2.54	1.48**	93.92	68.16
Mott F1	2.60	0.86	98.82	86.50
Cuba 115	2.01	0.48**	88.65	66.85
Cuba 169	2.08	0.44**	93.95	69.29
Cameroon	4.22	1.29	88.27	138.47
Napierzinho	4.00	0.69*	91.46	134.20
IJ 7125	4.05	1.12	97.20	135.89
IJ 7126	4.33	1.08	98.42	145.40
IJ 7127	1.80	1.49**	68.04	39.19
IJ 7136	4.05	1.07	90.96	134.79
Australiano	3.51	1.29*	97.20	111.27
13 AD	3.09	1.40**	93.45	95.08
10 AD	4.38	1.19	99.04	146.62
12 AD	2.02	1.67**	85.71	43.56
HV 290 23a x Elefante	1.54	0.49**	87.57	48.93
02 AD	3.44	0.97	99.99	115.55
08 AD	4.56	1.24	99.21	153.13

\* and \*\* significant at 5% and 1% of probability, respectively, by *t* test.

## Conclusions

There is variability between elephant grass genotypes to the tolerance of different aluminum levels, considering PSPA, PSR, APA and CR characteristics.

The level 15 ppm of aluminum was considered the most appropriated to identify materials tolerant and sensitive to aluminum toxicity.

PSPA and APA characteristics were the most suitable to future evaluation of elephant grass tolerance to aluminum, in nutrient solution.

Genotypes Taiwan A 121 and Australiano are adapted and stable by the evaluation parameters of the two methodologies used.

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