

Weeping lovegrass yield and nutritional quality provides an alternative to beef cattle feeding in semiarid environments of Argentina

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- 1 Weeping lovegrass yield and nutritional quality provides an
- 2 alternative to beef cattle feeding in semiarid environments of
- 3 Argentina
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- 14 **Abstract**
- Weeping lovegrass is a perennial warm-season grass spread over tropical and subtropical
- 16 regions worldwide. In Argentina, it has potential to colonize marginal production areas.
- 17 Therefore, the nutritional quality and yield of seven cultivars of weeping lovegrass was
- evaluated in a field trial located at Cabildo (Argentina) during two growing seasons. A
- 19 CRBD including five cultivars (Tanganyika, Morpa, Don Pablo, Don Juan, Don
- 20 Eduardo), two accessions (UNST9355 and 9446) and three blocks was used. Agronomic,
- 21 morphological and nutritional traits including fresh weight, dry weight, leaf length and

crown diameter, crude protein content, *in vitro* dry matter digestibility (IVDMD), neutral detergent fiber, acid detergent fiber and lignin content were determined. Two clippings per season were performed. The highest yields for winter growth and summer regrowth were obtained from Don Pablo and UNST9355, and Don Juan and UNST9446 respectively. Weeping lovegrass yield suggested that hexaploid cultivars were more productive than tetraploid ones under drought conditions. Also, IVDMDs indicated that digestibility decreased from winter to summer with the highest values obtained from Don Juan under drought conditions. Hence, breeding programs could select suitable parents from hexaploid cultivars such as Don Pablo and UNST9446 to be used in arid environments, while tetraploid cultivars such as Tanganyika and Morpa could be used in environments with less water restrictions.

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Abbreviattions

- 35 FMY, fresh matter yield; DMY, dry matter yield; MLL, maximum leaf length; CD,
- 36 crown diameter; FP, flowering percentage; INNP, inflorescence number per plant;
- 37 IVDMD, in vitro dry matter digestibility; CPC, crude protein content; NDF, neutral
- detergent fiber; ADF, acid detergent fiber; LC, lignin content.

Introduction

- Weeping lovegrass [Eragrostis curvula (Schrad.) Nees] is a perennial warm-season grass
- 41 mostly used as forage to support beef cattle production in arid and semiarid regions
- worldwide. This bunchgrass has been used as basis of pure or mixed pastures to support
- extensive cow-calf operations through spring and summer in marginal productivity areas

from countries such as United States and Argentina. Introduced in both countries in the
early '30s, this south African grass was spread from Oklahoma to Texas in the '70s
covering approximately 120,000 ha (Voigt et al., 2004) and now is found all throughout
the south of US according to the Plants Database (NRCS and USDA, 2011). Because of
weeping lovegrass easy domestication and dispersal, this pasture covered 700,000 ha
mostly represented by tetraploid apomictic genotypes in the '90s and it has the potential
to colonize over 5,000,000 ha in Argentina (Covas, 1991a). A recent survey indicated
that perennial pastures cover 8,643,100 ha distributed in central and southeast regions of
the country and, from those pastures, weeping lovegrass and wheatgrass (Elymus repens)
represent 5.4% and 6% becoming the second pure pastures behind alfalfa pastures
(INDEC, Encuesta Nacional Agropecuaria, 2002).
Weeping lovegrass is tolerant to a wide range of soils including light-textured and poor
soils with a wide pH range. Because of its physiological and morphological advantages,
that include well-developed root systems and epicuticular waxes, fast leaf rolling and fast
stomata closure responses, it is resistant to drought and extreme temperatures (Busso and
Brevedan, 1991; Echenique and Curvetto, 1991; Sanchez and Brevedan, 1991). Similarly
to other subtropical grasses, it has higher photosynthetic efficiency rate under high
temperature, light intensity and CO ₂ conditions due to its C4 metabolism. This efficiency
is translated to higher nutrient use efficiency and water use efficiency rates that are
reflected by faster responses to nitrogen fertilization and watering. In this sense, minimal
management requirements will increase growth rate and yield (Busso and Brevedan,
1991; Laborde, 1991; Sanchez and Brevedan, 1991). Therefore, weeping lovegrass seems
to be one of the most promising candidates to colonize these marginal areas in the center

67	and south east of Argentina where beef cattle production has been displaced by soybean
68	production in the last few years.
69	To improve nutritional quality of forage grasses, breeding programs have different
70	objectives including increasing voluntary intake (VI), dry matter yield (DMY), in vitro
71	dry matter digestibility (IVDMD), crude protein content (CPC) and water soluble
72	carbohydrate levels (WSC), and decreasing lignin and alkaloid contents (Wang et al.,
73	2001; Wilkins and Humphreys, 2003). Other breeding strategies include to enhance plant
74	persistence, tolerance to environmental stresses, resistance to insect pests and viral and
75	fungal diseases, and to increase seed yield. Because of most of these traits are
76	quantitative, breeding programs focused on phenotypic selection and progeny tests
77	including full-sib or half-sib family selection of crosspollinated grasses. Basically, traits
78	such as DMY and IVDMD with broad-sense heritabilities ranking between 30-70%,
79	showed an increase of 10 % decade (Wilkins and Humphreys, 2003). Other classical
80	breeding approaches such as gene introgression by backcrossing and chromosome
81	doubling were reported with limited success. Weeping lovegrass breeding efforts were no
82	different from other grass breeding programs. Early on, superior genotypes were
83	introduced from Africa to the US and Argentina (Voigt et al., 2004). These genotypes
84	included tetraploid cultivars that were successively crossed and produced highly
85	apomictic hybrid progenies checked by progeny tests. Slow progress was made in these
86	crosses because hybrid vigor and IVDMD were negatively correlated to sexuality and
87	winterhardiness respectively (Voigt, 1984). Furthermore, these hybrids were highly
88	apomictic and they could not reach commercial levels of seed production (Voigt et al.,
89	2004). However, later studies showed that yield-related traits such as dry matter, crown

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diameter and leaf length were highly stable through successive seasons although they varied through different environments indicating a strong interaction between genotypes and years (Di Renzo et al., 2000; Ibañez et al., 2001). Therefore, further efforts were focused on indirect selection strategies that look for higher hereditability values and genetic correlation coefficients among different locations to determine which will be the best locations to evaluate these agronomic traits (Di Renzo et al., 2003). Recently, new breeding efforts are developed at the Agronomy Department of the Universidad Nacional del Sur, where simultaneously to this study, crosses between sexual and apomictic tetraploids were made (OTA-S – PI 574506-vs. Tanganyika – PI 234217 -, Meier et al., unpublished) and a further evaluation of the agronomic and reproductive traits of the hybrid progeny will identify those hybrids with potential to become new cultivars. Therefore, our objectives were to characterize different weeping lovegrass cultivars by evaluating their agronomic, morphological and nutritional traits with the purpose of selecting those superior genotypes for their further inclusion in traditional and/or molecular breeding programs or biotechnological research projects.

Materials and Methods

Experimental field location and environmental conditions

This field experiment was conducted at the Experimental Station of the Asociación de Cooperativas Argentinas (ACA) located at Cabildo County, Buenos Aires, Argentina (39° 36' S, 61° 64' W) during two consecutive years (2008-2009 and 2009-2010). Soils are petrocalcic haplustoll and have a sandy loam texture with a calcareous hardpan layer at 50 cm (Rosell et al., 1992). Temperature and precipitation data were provided by a

- 112 meteorological station Davis Weather Monitor II (Davis Instruments Corp., Hayward,
- 113 CA) located at the experimental site. Also, historical averages for annual temperature and
- rainfall were obtained from the SMN (Servicio Metereológico Nacional, 2010).

Weeping lovegrass germplasm sources

- Weeping lovegrass cultivars Morpa and Tanganyika (apomictic tetraploids), and Don
- Pablo, Don Eduardo and Don Juan (apomictic hexaploids) were used. Also, two new
- accessions recently developed by somaclonal variation in our laboratory, UNST9446 and
- 119 UNST9355, were included. These accessions are the apomictic hexaploid progeny
- derived from a somaclonal variant obtained by anther culture from cv. Tanganyika (Polci,
- 121 2000). Specifically, the accession UNST9446 is registered as cv. Don Luis at the Instituto
- 122 Nacional de Semillas (INASE # RC9191/2006-2026).

Experimental design

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- The field trial was established in October 12th, 2002 (Polci, 2000). The experiment
- 125 consisted in a complete randomized block design (CRBD) including seven weeping
- lovegrass cultivars or accessions and three blocks. Plots were formed by four rows of
- eight plants per row with 0.5 m row spacing and 0.3 m space between plants. To avoid
- border effects, only the eight central plants from the two central rows were considered as
- a plot, and therefore measured and sampled. To determine winter and summer growth,
- two clippings were performed at November (November 20th, 2008 and November 5th,
- 2009) and April respectively (May 6th, 2009 and April 12th, 2010). Before spring growth
- and after a small rain, the field trial was fertilized with 100 kgN.ha⁻¹ applied as urea (46-

0-0) and supplementary watering was provided at the end of the summer (25 mm in
February, 2009, and January, February and March, 2010).
Morphological traits such as maximum leaf length (MLL), crown diameter (CD) and
percentage of flowering (FP) were measured during both years, while the number of
inflorescences or panicles per plant (INNP) was only measured during the second year.
Plots were hand clipped at 5-10 cm, agronomic traits such as FW and DW -after drying at
65C to constant weight- were determined, and fresh and dry matter yields were estimated
(FMY and DMY, respectively). After drying at 105C, subsamples were ground with a
2mm screen in a Wiley mill and used to determine nutritional traits including dry matter
(DM), ashes (A), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber
(ADF), lignin content (LC) and in vitro dry matter digestibility (IVDMD) by the Animal
Nutrition Laboratory of Facultad de Ciencias Agrarias from Universidad de Buenos
Aires (FAUBA).

Statistical analysis

To determine the fitness of the best model, agronomic and nutritional traits were evaluated by using the mixed model analysis in R (R Development Core Team, 2010) and InfoSat (Di Rienzo et al., 2010). Originally, cultivars, clipping dates and cultivars*clipping dates interaction were considered as factors with fixed effects and blocks or replicates as factors with random effects. However, the cultivar*clipping date interactions were significant for most agronomic and nutritional trait data (P≤0.05) (Supplemental Table 1). Therefore, these traits were analyzed using a mixed model with cultivars and blocks -as factors with fixed and random effects, respectively- for each clipping date. Also, to compare cultivar means within each clipping date a Least Square

- Difference (LSD) test was performed using InfoStat and R (P≤0.05) (Di Rienzo et al.,
- 157 2010; R Development Core Team, 2010).

Results

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Environmental conditions and weeping lovegrass production

160 Weeping lovegrass growth and development were affected by stressful environmental 161 conditions including freezing temperatures and drought during these two growing seasons 162 (Figure 1). According to average temperature data, plant growth was stimulated by 22.0 163 and 19.7C average temperatures through first and second summer periods (i.e. 164 November, 2008 to April 2009, and November, 2009 to April, 2010 respectively). 165 Instead, plant regrowth was delayed because average temperatures decreased to 11.2 and 166 12.1C through first and second winter periods (i.e. May to October, 2009 and 2010 167 respectively). In addition to this, there were at least 5 days of freezing temperatures per 168 month during five months –since May to Setember, 2008-, while the days with freezing 169 temperatures came two months later the next year –since July to October, 2009- (Figure 170 1A). These data indicated that the first winter was more stressful than the second one 171 because it had 28 days with freezing temperatures evenly distributed through the season 172 while the second winter only had 16 freezing days that came later in the season (Figure 173 1A). Also, weeping lovegrass growth was differentially affected by rainfall during these 174 two growing seasons (Figure 1B). Total rainfall reached 407.2 and 527.8 mm in the first 175 and second growing seasons respectively. These precipitations were evenly distributed 176 with 42.1 and 57.9% in the winter and summer of the first season, while they were 177 unevenly distributed with 15.1 and 84.9% in the winter and summer of the second season.

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According the Servicio Metereológico Nacional (SMN, 2010. URL http://www.smn.gov.ar), the decade averages corresponding to seasons 2008-2009 and 2009-2010 were 648 and 645 mm respectively, and rainfall was almost evenly distributed with 38 and 62% of the accumulated rainfall during winter and summer in both years. Therefore, precipitation data indicated that the first growing season occurred through a drier year with evenly distributed rainfall while the second growing season occurred through a year with more rainfall but unevenly distributed between a drier winter and a rainy summer periods (Figure 1B). These environmental conditions were directly reflected on overall weeping lovegrass biomass production (Figure 1C). In this sense, FMY and DMY were estimated in 7,118.8 and 4,264.9 kg.ha⁻¹ during the first growing season, and 19,114 and 9,993.8 kg.ha⁻¹ during the second growing season. Moreover, these predicted yield values were almost evenly distributed between winter and summer at the first growing season (with 53.46 and 56.67% and 46.54 and 43.32 % respectively) while they were unevenly distributed at the second growing season (with 8.76 and 9.42%) during the winter and 91.24 and 90.58% during the winter respectively) (Figure 1C).

Agronomic and Morphological traits

Weeping lovegrass cultivar growth was evaluated through four different clipping dates representing winter regrowth and summer growth at the first growing season (by November 20th, 2008 and May 6th, 2009), and winter regrowth and summer growth at the second growing season (by November 5th, 2009 and April 12th, 2010). In first place, the highest FMY and DMY were produced by cv. Don Pablo with 4,524.3 and 2,857.7 kg.ha ⁻¹ respectively. Although these values were significantly different from 3,344.3 and 2,133.1 kg.ha ⁻¹ obtained by cv. Morpa at the first clipping date. In second place, the

highest FM and DMYs were 4,866.6 and 2,7712.0 kg.ha ⁻¹ , and they were produced by
UNST9446. These values were significantly higher than those values estimated for cv.
Tanganyika, while they were not significantly different from those produced by cv. Don
Pablo. In third place, cv. Don Pablo also produced the highest FM and DMYs with
2,033.6 and 1,139.5 kg.ha ⁻¹ . These values did not differ from those values obtained by
UNST9355, but they differed significantly from those values obtained by cv. Don
Eduardo at the third clipping date. In fourth place, cv. Don Juan produced the highest
FMY and DMY with 23,808.5 and 12,070.6 kg.ha ⁻¹ , followed by cvs. Don Pablo,
Tanganyika and Morpa; and these yields were significantly higher than those values
calculated for cv. Don Eduardo at the fourth clipping date. In this sense, it is important to
notice that these values were the highest FM and DMYs estimated among different
clipping dates (Figure 2).
Maximum leaf length and CD were conserved through both growing seasons (Figure 3).
According to MLL data, UNST9446 showed a more upright and open growing habit with
longest leaves ranking between 43.4 and 46 cm length in the first and second winter and
between 45.5 and 110 cm in the first and second summer respectively. While cv. Don
Juan showed a postrate growing habit with shortest leaves ranking between 28.9 and 27.4
cm during the first and second winter and between 30.5 and 100 cm during the first and
second summer. However, MLL values from UNST9446 and cv. Don Pablo were not
significantly different during both winters indicating that both genotypes had faster leaf
growth rates during these periods; and cv. Don Juan and Tanganyika did not differed
significantly in the second summer indicating that both cultivars had slower leaf growth
rates during a summer without climate restrictions (Figure 3). Also, weeping lovegrass

cultivars showed a small or null plant growth rate reflected in a small variation of the CD
values (Figure 3). Specifically, plants from cv. Don Eduardo showed the highest CD
values, while plants from UNST9355 were significantly smaller during the first season
(with 21.4 and 20.1 cm at the first clipping date, and 19.1 and 17.7 cm after the second
clipping date, respectively). These CDs decreased from winter to summer probably
indicating partial plant death due to drought and winter freezing temperatures. Although,
cultivar and accessions plant growth rates were similar and increase from winter to
summer during the second season with cvs. Don Juan and Morpa reaching the largest CD
values (21.5 and 21.9 cm respectively)(Figure 3).
Weeping lovegrass flowers throughout spring and summer, beginning in early Setember
and ending in late April or May, therefore it is possible to detect those cultivars that
respond early on, at the end of the winter, and those cultivars that keep flowering as long
as warm days last, at the end of the summer or beginning of the fall (Figure 4). Thus,
UNST9446 and hexaploid cultivars including Don Pablo, Don Eduardo and Don Juan
(with 100 and 95% FP respectively) flowered significantly earlier than UNST9355 and
tetraploid cultivars Tanganyika and Morpa (with 67, 71 and 50% FP respectively) at the
first clipping date. Although, only UNST9446 and cv. Don Juan kept flowering at the
beginning of the fall (with 96 and 100% FP respectively) while cultivar flowering was
significantly reduced in cvs. Don Eduardo and Tanganyika (with 67 and 63%F
respectively) at the second clipping date. Also, UNST9355 and cvs. Don Pablo and Don
Juan flowered early (with 100, 88 and 96% FP) while cv. Don Eduardo flowering was
significantly delayed (17% FP) at the third clipping date; while cultivar flowering was
steady all throughout the season in most cultivars except by cy. Tanganyika that showed

a reduced flowering (71% FP) at the fourth clipping date. Moreover, UNST9355 and cvs. Don Juan and Don Pablo showed a 4.75, 4.33 and 4.83 INNP respectively, which was significantly higher than 0.3 INNP observed in cv. Don Eduardo indicating an early flowering at the third clipping date, that increased through the season reaching 72.4 INNP in cv. Don Juan, 20 to 27 INNP in the other tetraploid and hexaploid cultivars and 12.6 INNP in cv. Don Eduardo at the fourth clipping date (Figure 4).

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Nutritional traits

Most forage grasses are characterized by nutritional traits that increase and/or decrease their nutritive value, and these traits include CP and IVDMD as traits which improve nutritional quality and NDF, ADF and LC as traits that reduce their nutritional quality (Figure 5). Because of plant developmental and seasonal growth, IVDMD varies reaching the highest values during winter regrowth when vegetative growth is reduced and therefore nutritional quality is higher. Thus, IVDMD percentages varied between 58.3-58.0% and 53.6% from cvs. Morpa and Don Juan, and from cv Tanganyika as the highest and lowest percentages obtained after the first winter regrowth while IVDMD percentages varied between 45.3 and 38.1% IVDMD from cvs. Don Juan and Morpa after summer growth and flowering. Moreover, this tendency was reinforced by a drier winter and a rainy summer during the second season, and translated to higher IVDMD values after winter regrowth (ranking between 68.7% and 61.1 to 64.4% from cv. Don Juan and the remaining cultivars respectively) and IVDMD values still lower than those values obtained from the previous summer growth (varying between 47.9 and 44.1% from cv. Don Eduardo and UNST9446 respectively) (Figure 5A). Also, CPC followed these

patterns through both growing seasons (Figure 5A). Therefore, CP content varied
between 6.8% from tetraploid cultivars and 5.6% from cv. Don Eduardo after the first
winter and between 10.9 from cv. Don Eduardo and 9.6-9.8% from UNST9446 and cv.
Don Pablo respectively after the second winter growth. Although, these CP contents
decreased after both summers ranking between 7.4 and 8.9% from UNST9446 and cv.
Don Eduardo after the first summer and 4.4 and 3.3% from cv. Tanganyika and
UNST9446 during the second summer respectively (Figure 5A).
It is important to recall that biomass digestibility and degradability are mainly affected by
plant cell wall components including cellulose, hemicelluloses and lignin. Therefore,
high cell wall content -indirectly detected by traits such as NDF, ADF and LC- reduces
IVDMD and Dry Matter Intake (DMI) which directly decreases animal performance
(Figure 5B). In this sense, NDF and ADF percentages decreased from winter regrowth to
summer growth during the first season but decreased further with the second winter and
increased furthermore with the second summer. So that, no significant differences were
observed among NDF percentages from different cultivars in the first winter regrowth
(with 72.5 and 73.8% from cvs. Don Pablo and Tanganyika respectively). Although,
NDF percentages from UNST9446 and tetraploid cultivars were significantly different
from the NDF percentage obtained from cv. Don Eduardo after summer growth (71.3-
71.5% vs. 68.7% respectively). However, NDF percentages were lower after second
winter regrowth (with 65.4 and 70.8% from cvs. Don Juan and Morpa) and these NDF
percentages increased after summer growth reaching between 74.3-74.6 and 76.8% from
UNST9446, 9355 and cv. Don Juan, and cv. Morpa, respectively (Figure 5B). Also, ADF
percentages varied between 34.5 and 38.3% from cvs. Morpa and Don Juan after the first

winter regrowth, but these values decreased further after the first summer reaching between 32.4 and 35.3% for cvs. Don Eduardo and UNST9446 respectively. Although, the lowest ADF percentages were reached during the second winter ranking between 30.4 and 34.0% for cv. Don Juan and UNST9355 respectively, while the highest ADF percentages were obtained after the second summer growth ranking between 38.0-38.7% by UNST9446 and hexaploid cultivars respectively and 36.0% by cv. Tanganyika (Figure 5B). On the other hand, LC showed almost null and small variation through the first and second growing seasons respectively (Figure 5B). Thus, LC varied between 3.6 and 4.7% from cvs. Don Pablo and Don Juan at the first clipping date; and between 3.9% from cv. Don Eduardo and UNST9355, and 4.5% from cvs. Don Juan and Morpa at the second clipping date. However, LC shifted from 2.8-4.0% for cv. Don Pablo and UNST9446, and cv. Tanganyika respectively at the third clipping date, to 4.0-4.8% from cvs. Don Eduardo and Don Juan at the fourth clipping date (Figure 5B).

Discussion

Weeping lovegrass [Eragrostis curvula (Schrad.) Nees] is one of the most important forage grasses supporting extensive beef cattle production in mixed pastures through arid and seamiarid regions from Argentina. In this sense, it has been used as a complement of native pastures and it performs an important role avoiding desertification and helping to native pasture restoration in these regions (Covas, 1991a; Guevara et al., 2005). Because of its efficient biomass production under restrictive environments, it could be used for grazing through the summer and as standing deferred forage through the winter (Covas, 1991a; Covas, 1991b; Gargano et al., 2001; Stritzler et al., 1996). An early comparative study on forage grass nutritional quality and yield, mostly focused on winter yield,

established five clipping dates and compared four grasses including weeping lovegrass
cv. Tanganyika, switchgrass cv. Pathfinder (Panicurn virgutum L.), kleingrass (Panicurn
colorutum L.) and robies cocksfoot (Tetrachne dregei) during years 1991 and 1992
(Stritzler et al., 1996). In this study, authors showed that cv. Tanganyika produced a
higher DMY (i.e. 10,132 kg.ha ⁻¹) with a slightly lower nutritional quality. In contrast to
these authors, we observed that cv. Tanganyika DMY was significantly lower -reaching a
10-fold yield decrease in the second winter -, while nutritional quality was higher through
both winters. However, these DMY and nutritional quality differences could be partially
explained by the number of clippings done through the winter. While Stritzler et al
(1996) performed 5 successive clippings stimulating a continuous vegetative growth that
reduced nutritional quality, we only performed one clipping through the winter. Similarly
to our study, Gargano et al. (2001) evaluated DMY and IVDMD of two warm-season
grasses -weeping lovegrass cv. Tanganyika and pangolagrass (Digitaria erianthra) cv.
Irene- where they carried out two clipping dates by growing season during four
consecutive years (at October 15 th and February 20 th , 1995, 1996, 1997 and 1999). Here,
authors observed that DMY and IVDMD differential patterns were conserved through
successive growing seasons in accordance to our results. They observed that both grasses
produced 80% DMY through summer growth -similarly to our second growing season-
and suggested that weeping lovegrass higher yield is given by an earlier winter regrowth,
higher winterhardiness and less lodging Specifically, cv. Tanganyika DMY reached
3,591 and 729 kg.ha ⁻¹ -with 9.2 and 3.9% CPC, and 42 and 50% IVDMD in summer
growth and winter regrowth respectively- limiting animal performance because it
provides only 50% of daily requirement to assure normal cattle weight gain during winter

(Gargano et al., 2001). Later on, Gargano et al. (2006) performed other comparative
study evaluating three warm-season grasses -weeping lovegrass, pangolagrass and wool
grass (Anthephora pubescens) cv. Woollie- and their performance under three
fertilization rates during spring growth over two consecutive years (2001-2002 and 2002-
2003). They reported that cv. Tanganyika produced intermediate DMYs and CPC
depending on the fertilization rate (Gargano et al., 2006). However, these comparative
studies were performed across species and there are no further studies on cultivar
potential within these forage grasses. Here, it is important to recall that within weeping
lovegrass there are several agronomic types, including curvula, robusta and chloromelas
types among others, that were previously described (Leigh, 1960; Voigt, 1991).
Therefore, we evaluated not only those cultivars that are commercially available -
tetraploid curvula-type cultivars such as Morpa and Tanganyika- but also other promising
candidates including hexaploid cultivars like Don Pablo and Don Eduardo with a
robusta-type, and Don Juan with a chloromelas-type; and two accessions -UNST9446
and UNST9355- generated in our laboratory. We observed that weeping lovegrass
cultivar yields, expressed as FMY and DMY, were restricted by environmental
conditions through the first three clipping dates while these conditions were favorable
and stimulated cultivar growth reaching an average 10-fold yield increase at the fourth
clipping date. However, cultivars and accessions with intermediate yields showed
differential patterns indicating that hexaploid cultivars from robusta or chloromelas types
and UNST9446 are more efficient than tetraploid curvula-type cultivars such as
Tanganyika and Morpa under mild winters and summers. Also, MLL and CD - reflecting
indirectly leaf growth and plant growth rates- indicated that vegetative growth was

reduced through the first season, and almost null through the second winter, while it fully
recovered through the second summer. Moreover, vegetative growth translated into
reproductive growth, indirectly measured by FP and INNP, indicating cultivar potential
for seed production which is an important trait when accessions are considered for
commercial purposes. Generally, nutritional traits such as IVDMD and CPC are
negatively correlated to FM and DMYs while traits such as NDF, ADF and LC are
positively correlated to these yields because while vegetative and reproductive plant
growth advance, cell wall components increase due to secondary growth and flowering.
Nowadays, there are no breeding programs to improve weeping lovegrass yield or
nutritional quality in Argentina. Although, early breeding efforts indicated that yield was
correlated to MLL, CD and DM with 0.86, 0.84 y 0.84 repeatability values in a
population with 18 weeping lovegrass hybrids showing that 98% accuracy could be
reached with only a two-year study (Di Renzo et al., 2000; Voigt et al., 1996). Further
studies concluded that these traits were highly variable and, environment and
genotype*environment interactions were significant factors explaining 65 and 14.5%
variation respectively (Ibañez et al., 2001). Therefore, recent breeding efforts were
focused on evaluating indirect selection efficiency in three locations over two years
adding up to six environments. Di Renzo et al. (2003) observed that indirect selection
efficiency found in Bahía Blanca was no different from selection efficiency found in Río
Cuarto, and that DM heritabilities found in Bahía Blanca and Villa Mercedes were higher
allowing a faster genetic advance for future breeding programs in these locations.
Nevertheless, weeping lovegrass nutritional quality was not included in these studies. So
far, this is the first report focused on evaluating not only agronomic and morphological

but also nutritional traits of seven different weeping lovegrass sources –including two new materials and 5 cultivars- in semiarid regions of Argentina. We observed that cv. Tanganyika -the most widely grown and studied cultivar- DMY was lower than hexaploid cultivar and UNST9446 DMYs under stressful conditions like those observed through the first growing season and the second winter. On the other hand, it ranked better than them when the environmental conditions were favorable. Hence, it will be possible to select suitable parents for breeding programs with bidirectional selection where cultivars such as hexaploid cultivars like Don Pablo and UNST9446 will be used in arid environments while tetraploid cultivars such as Tanganyika and Morpa will be used in environments with less water restrictions.

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469	
470	Legends
471	Figure 1. Weeping lovegrass biomass production affected by environmental conditions
472	through two successive growing seasons (April, 2008-2009 and 2009-2010). A)
473	Temperature effects expressed as maximum, average and minimum values and as number
474	of days with freezing temperatures, B) Rainfall effects registered as monthly rainfall and
475	relative humidity. All these data were obtained from a meteorological station located at
476	the field site. The last decade monthly average rainfall was provided by the SMN (2010).
477	C) Overall weeping lovegrass production estimated as Fresh Matter Yield and Dry Matter

478	Yield for the four clipping dates. Different letters indicate significant differences among
479	accessions and cultivars by LSD test (P≤0.05).
480	
481	Figure 2. Weeping lovegrass yield measured through four different clipping dates during
482	two successive growing seasons (April, 2008-2009 and 2009-2010). Fresh Matter Yield
483	and Dry Matter Yield (FMY and DMY respectively) estimated as kg. ha -1 for two
484	accessions (UNST9446 and 9355) and five different cultivars (Tanganyika, Morpa, Don
485	Pablo, Don Juan and Don Eduardo). Different letters indicate significant differences
486	among accessions and cultivars by LSD test (P≤0.05).
487	
488	Figure 3. Weeping lovegrass morphological traits measured through four different
489	clipping dates during two successive growing seasons (April, 2008-2009 and 2009-2010).
490	Maximun Leaf Length and Crown Diameter (MLL and CD) registered for two accessions
491	(UNST9446 and 9355) and five different cultivars (Tanganyika, Morpa, Don Pablo, Don
492	Juan and Don Eduardo). Different letters indicate significant differences among
493	accessions and cultivars by LSD test (P≤0.05).
494	
495	Figure 4. Weeping lovegrass flowering traits measured through four different clipping
496	dates during two successive growing seasons (April, 2008-2009 and 2009-2010).
497	Flowering Percentage and Inflorescence Number per Plant (FP and INNP respectively)
498	registered for two accessions (UNST9446 and 9355) and five different cultivars
499	(Tanganyika, Morpa, Don Pablo, Don Juan and Don Eduardo). Different letters indicate
500	significant differences among accessions and cultivars by LSD test (P≤0.05).

501	
502	Figure 5. Weeping lovegrass nutritional traits measured through four different clipping
503	dates during two successive growing seasons (April, 2008-2009 and 2009-2010). A) In
504	Vitro Dry Matter Digestibility and Crude Protein Content (IVDMD and CPC
505	respectively), B) Neutral Detergent Fiber, Acid Detergent Fiber and Lignin Content
506	(NDF, ADF and LC respectively) registered for two accessions (UNST9446 and 9355)
507	and five different cultivars (Tanganyika, Morpa, Don Pablo, Don Juan and Don
508	Eduardo). Different letters indicate significant differences among accessions and cultivars
509	by LSD test (P≤0.05).
510	
511	Supplementary Table 1. Mixed Model analyses for agronomic, morphological and
512	nutritional traits of weeping lovegrass [Eragrostis curvula (Schrad.)Nees] . These results
513	correspond to the model where agronomic morphological and nutritional traits = fixed
514	effects (cultivar+clipping date+ cultivar*clipping date) + random effects (blocks) with
515	the FMY, DMY, MLL, CD and FP sequential hypotheses tested with N=664; intercept
516	fd=1, cultivar fd=6, clipping dates fd=3 and cultivar*clipping date fd=18, and
517	Fvalue=634; the INNP sequential hypotheses tested with N=323; intercept df=1, cultivar
518	fd=6, clipping dates df=1, cultivar*clipping date df=6, and Fvalue=313; and the IVDMD,
519	PC, NDF, ADF, LC sequential hypotheses tested with N=84, intercept df=1, cultivar
520	df=6, clipping dates df=3 and cultivar*clipping date df=18, and Fvalue=54.
521	
522	

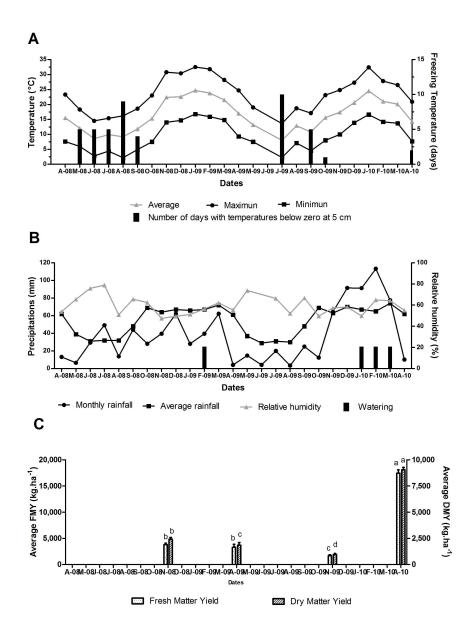
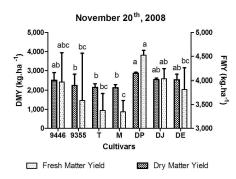
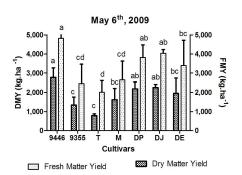
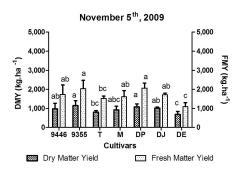


Figure 1. Weeping lovegrass biomass production affected by environmental conditions through two successive growing seasons (April, 2008-2009 and 2009-2010). A) Temperature effects expressed as maximum, average and minimum values and as number of days with freezing temperatures, B) Rainfall effects registered as monthly rainfall and relative humidity. All these data were obtained from a meteorological station located at the field site. The last decade monthly average rainfall was provided by the SMN (2010). C) Overall weeping lovegrass production estimated as Fresh Matter Yield and Dry Matter Yield for the four clipping dates. Different letters indicate significant differences among accessions and cultivars by LSD test (P≤0.05).

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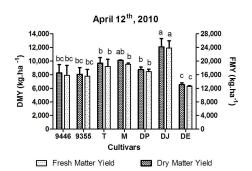
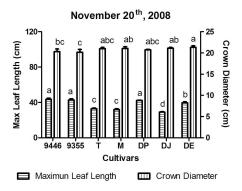
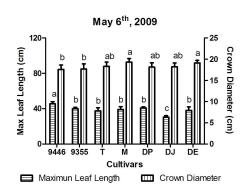
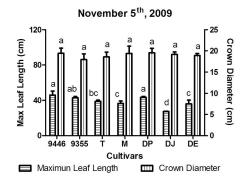


Figure 2. Weeping lovegrass yield measured through four different clipping dates during two successive growing seasons (April, 2008-2009 and 2009-2010). Fresh Matter Yield and Dry Matter Yield (FMY and DMY respectively) estimated as kg. ha -1 for two accessions (UNST9446 and 9355) and five different cultivars (Tanganyika, Morpa, Don Pablo, Don Juan and Don Eduardo). Different letters indicate significant differences among accessions and cultivars by LSD test ($P \le 0.05$). $192 \times 198 \, \text{mm}$ (300 x 300 DPI)







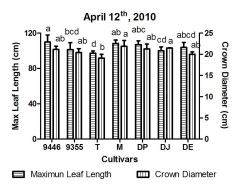
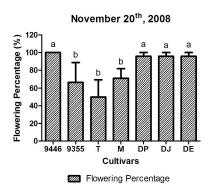
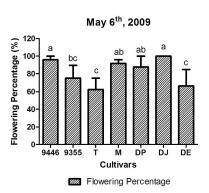


Figure 3. Weeping lovegrass morphological traits measured through four different clipping dates during two successive growing seasons (April, 2008-2009 and 2009-2010). Maximun Leaf Length and Crown Diameter (MLL and CD) registered for two accessions (UNST9446 and 9355) and five different cultivars (Tanganyika, Morpa, Don Pablo, Don Juan and Don Eduardo). Different letters indicate significant differences among accessions and cultivars by LSD test ($P \le 0.05$). $189 \times 203 \text{mm} (300 \times 300 \text{ DPI})$





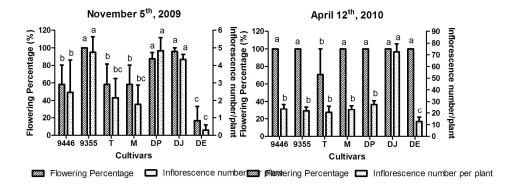


Figure 4. Weeping lovegrass flowering traits measured through four different clipping dates during two successive growing seasons (April, 2008-2009 and 2009-2010). Flowering Percentage and Inflorescence Number per Plant (FP and INNP respectively) registered for two accessions (UNST9446 and 9355) and five different cultivars (Tanganyika, Morpa, Don Pablo, Don Juan and Don Eduardo). Different letters indicate significant differences among accessions and cultivars by LSD test (P≤0.05).

201x207mm (300 x 300 DPI)

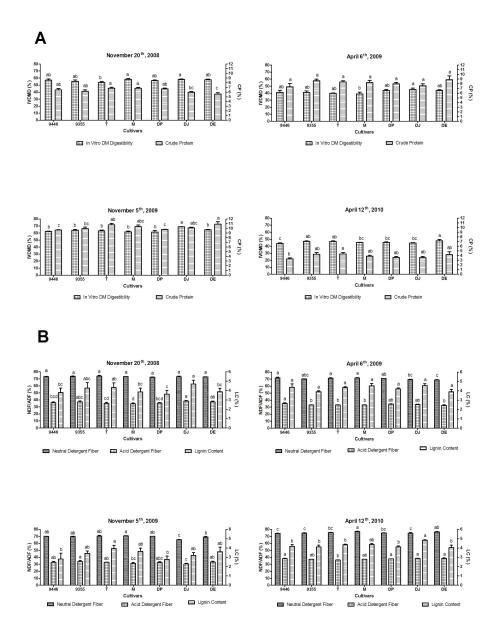


Figure 5. Weeping lovegrass nutritional traits measured through four different clipping dates during two successive growing seasons (April, 2008-2009 and 2009-2010). A) In Vitro Dry Matter Digestibility and Crude Protein Content (IVDMD and CPC respectively), B) Neutral Detergent Fiber, Acid Detergent Fiber and Lignin Content (NDF, ADF and LC respectively) registered for two accessions (UNST9446 and 9355) and five different cultivars (Tanganyika, Morpa, Don Pablo, Don Juan and Don Eduardo). Different letters indicate significant differences among accessions and cultivars by LSD test (P≤0.05).

197x250mm (300 x 300 DPI)

Sequential	Intercept		Cultivar		Clipping date		Cultivar*Clipping date	
Hypotesis tests	P-value	Significance	P-value	Significance	P-value	Significance	P-value	Significance
Agronomic traits								
FMY	279.15	***	6.27	***	605.18	***	5.36	***
DMY	395.53	***	5.39	***	542.12	***	4.84	***
MLL	25381.83	***	27.31	***	2292.55	***	2.67	**
CD	854.33	***	2.69	**	34.24	***	1.03	ns
FP	465.32	***	13.57	***	29.29	***	7.86	***
INNP	58.23	***	59.93	***	661.3	***	50.18	***
Nutritional traits								
IVDMD	37436.72	***	3.22	**	340.52	***	1.63	ns
CP	10781.8	***	3.12	**	379.29	***	1.26	ns
NDF	380986.51	***	6.84	***	128.03	***	2.94	**
ADF	4067.85	***	2.31	*	63.69	***	2.01	**
LC	460.85	***	2.83	**	16.83	***	1.03	ns

Supplementary Table 1.- Mixed Model analyses for agronomic, morphological and nutritional traits of weeping lovegrass [*Eragrostis curvula* Nees (Schrad.)] . These results correspond to the model where agronomic morphological and nutritional traits = fixed effects (cultivar+clipping date+ cultivar*clipping date) + random effects (blocks) with the FMY, DMY, MLL, CD and FP sequential hypotheses tested with N=664; intercept fd=1, cultivar fd=6, clipping dates fd=3 and cultivar*clipping date fd=18, and Fvalue=634; the INNP sequential hypotheses tested with N=323; intercept df=1, cultivar fd=6, clipping dates df=1, cultivar*clipping date df=6, and Fvalue=313; and the IVDMD, PC, NDF, ADF, LC sequential hypotheses tested with N=84, intercept df=1, cultivar df=6, clipping dates df=3 and cultivar*clipping date df=18, and Fvalue=54.