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Review Paper

Tropical forage legumes for environmental benefits: An overview

Leguminosas forrajeras tropicales para beneficios ambientales: Una sinopsis

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Abstract

Ruminant livestock production in the tropics, particularly when based on pastures, is frequently blamed for being detrimental to the environment, allegedly contributing to: (1) degradation and destruction of ecosystems, including degradation and loss of soil, water and biodiversity; and (2) climate change (global warming). In this paper we argue that, rather than being detrimental, tropical forage legumes can have a positive impact on the environment, mainly due to key attributes that characterize the Leguminosae (Fabaceae) family: (1) symbiotic nitrogen fixation; (2) high nutritive value; (3) deep-reaching tap-root system; (4) wide taxonomic and genetic diversity; and (5) presence of particular secondary metabolites. Although there are also potential negative aspects, such as soil acidification and the risks of introduced legumes becoming invasive weeds, we submit that legumes have potential to contribute significantly to sustainable intensification of livestock production in the tropics, along with the provision of ecosystem services. To further assess, document and realize this potential, research for development needs in a range of areas are indicated.

Keywords: Biodiversity, ecosystem services, GHG emissions, land rehabilitation, soil enhancement, symbiotic nitrogen fixation.

Resumen

La producción ganadera de rumiantes en el trópico, especialmente cuando es basada en pasturas, frecuentemente es considerada como perjudicial para el medio ambiente, ya que supuestamente contribuye con: (1) la degradación y destrucción de ecosistemas, incluyendo la pérdida de suelo, agua y biodiversidad; y (2) el cambio climático (calentamiento global). En el artículo se exponen argumentos para mostrar que, en lugar de ser perjudiciales, las leguminosas forrajeras tropicales pueden impactar positivamente en el medio ambiente, principalmente debido a sus atributos clave que son característicos de la familia de las Leguminosae (Fabaceae): (1) fijación simbiótica de nitrógeno; (2) alto valor nutritivo; (3) sistema de raíz pivotante profundo; (4) amplia diversidad taxonómica y genética; y (5) presencia de metabolitos secundarios particulares. Aunque se deben reconocer aspectos negativos como la contribución potencial a la acidificación del suelo y el riesgo de convertirse en malezas invasoras, concluimos que las leguminosas forrajeras tienen un potencial significativo para contribuir a la intensificación sostenible de la producción ganadera en el trópico, junto con la prestación de servicios ecosistémicos. Sugerimos una serie de áreas donde se requiere de investigación para evaluar más a fondo, documentar y realizar este potencial.

Palabras clave: Biodiversidad, emisiones de GEI, fijación simbiótica de nitrógeno, mejoramiento del suelo, rehabilitación de tierras, servicios ecosistémicos.

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Introduction

Feeding the world population is a major challenge for the future when one considers that in 2050 there will be an expected >9 billion people on this planet. Consequently, food production must be increased and intensified (FAO 2010). At the same time, there is growing concern about the environmental impact of agricultural production, in particular of livestock (Steinfeld et al. 2006). While past agricultural research focused primarily on increased production, it is now well recognized that ecological concerns must be addressed as well if environmentfriendly production strategies are to be developed and sustainable intensification (SI) is to be achieved (Garnett et al. 2013; The Montpellier Panel 2013). SI encompasses increased production from existing farmland without negatively affecting the environment, and the approach has been adopted as a policy goal for a number of national and international organizations working towards sustainable development goals. This SI policy goal applies also to research on tropical pastures and forages and is reflected, for example, in the theme of the last International Grassland Congress (New Delhi, India, November 2015): Sustainable Use of Grassland Resources for Forage Production, Biodiversity and Environmental Protection.

Two recent overview analyses of tropical forage-based livestock production systems vis-à-vis the environment and the need for SI concluded that such systems can have a positive impact on the environment (Peters et al. 2013; Rao et al. 2015). In tropical production systems, the term 'forages' refers mostly to grasses, since adoption of legume technology in the past has been rather low (Shelton et al. 2005). We hypothesize, however, that tropical forage legumes do have the potential to play a particular, positive role in addressing environmental concerns.

Therefore, complementing the above-mentioned overviews and in order to contribute to the development of research strategies, in this paper we analyze the effects of tropical forage legumes (pasture plants for grazing or fodder plants for cut-and-carry or browsing) on the environment. For this, we briefly: outline the main anthropogenic environmental issues; highlight some essentials related to livestock production and the environment; and discuss the key attributes of forage legumes that contribute to natural resource conservation and environmental protection with a particular emphasis on adaptation to and mitigation of climate change. We then examine the potential of tropical forage legumes to have a positive impact on environmental issues and provide ecosystem services.

Environmental issues

The main, human-induced environmental problems, as currently perceived, are related to: natural resources, including biodiversity; and climate change.

Regarding *natural resources*, it is generally accepted that the major issues are: (1) ecosystem destruction and degradation; (2) soil degradation and loss; (3) water degradation and loss; and (4) biodiversity degradation and loss. Obviously, these problem areas are all interrelated.

Regarding *climate change* and its major manifestations (global warming leading to modifications of rainfall regimes and both flooding and drought phenomena), <u>IPCC (2014)</u> states that the main driver is increased anthropogenic greenhouse gas (GHG) emissions, mainly carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O).

Livestock production and the environment: Some background essentials

When considering livestock production in the tropics and its impact on the environment, a few issues should be highlighted:

- In the scientific and non-scientific communities, livestock production, including grazing, is blamed for severe negative impacts on the environment (e.g. <u>Steinfeld et al. 2006; Hyner 2015</u>). Livestock production is estimated to contribute 14.5% of all anthropogenic GHG emissions globally (Gerber et al. 2013).
- The demand for animal products, especially foods derived from livestock, is expected to increase consid-erably in the future, particularly in South, East and Southeast Asia, and to a lesser extent in Sub-Saharan Africa, as a consequence of increasing living standards (Rosegrant et al. 2009; Robinson and Pozzi 2011).
- In view of physical limitations to expansion of land area for agricultural production (both crop and livestock), future production increases must come mainly from intensification of production systems (<u>The Montpellier Panel 2013</u>).
- Ruminant livestock (e.g. cattle, buffalo, sheep, goats) play an important role as they convert vegetation, which is unsuitable as food for humans, into high-quality products for human consumption. Nonetheless ruminants are fed grain-based diets (such as in feedlots), and this practice is in direct competition with humans for that food source (Mottet et al. 2017).
- Tropical grazing lands often occupy marginal land that is unsuitable or only marginally suitable for crop production, because of constraints imposed by soil

physical and chemical properties, topography (including slopes and waterlogging) and climatic conditions (Rao et al. 2015). Similarly, some lands are temporarily unsuitable for crop production due to drought or excess of water, and these areas are expected to increase in the tropics (Zabel et al. 2014). Here, crop production could benefit from a crop-forage rotation.

• As far as research on tropical pastures and forages is concerned, the literature provides almost no indication that, in the past, environmental issues have played a major role in forage development and utilization. Notable exceptions are the concerns expressed by McIvor et al. (1997; 2005) and Noble et al. (2000).

Key attributes of legumes

The main 5 features of this plant family in general are summarized as follows:

- 1. Legumes in the Papilionoideae subfamily and in what used to be the Mimosoideae subfamily [now the 'mimosoid clade' in the newly defined Caesalpinioideae subfamily (LPWG 2017)] and a few taxa in the Caesalpinioideae subfamily can fix, in symbiosis with rhizobia (Bradyrhizobium, Rhizobium), atmospheric nitrogen (N). Therefore they have the potential to: (1) be N self-sufficient; and (2) increase N availability in the soil for associated or subsequent crops, forage grasses and soil biota. Depending on legume species, effectiveness of rhizobium strains, nutrient supply (mainly phosphorus, potassium and the trace element molybdenum), climatic conditions and assessment method applied, published data for symbiotic N fixation (SNF) by tropical forage legumes cover a wide range, e.g. 15-158 kg N/ha/yr using ¹⁵N methodologies (Thomas 1995); a recent example is the range of 123-280 kg symbiotically fixed N/ha/yr in 6 Arachis glabrata cultivars, reported by Dubeux et al. (2017a). Total input of SNF to mixed grass-legume pasture systems can range from 98 to 135 kg N/ha/yr (Boddey et al. 2015). This attribute is particularly important in production systems that depend on external N inputs (Douxchamps et al. 2014).
- Most forage legumes have high nutritive value for ruminants, mainly in terms of concentration of crude protein (CP) (percentage N x 6.25) but also of energy (Lüscher et al. 2014). This feature can be particularly significant in mixtures with, or as complement to, grasses with CP levels often below livestock maintenance requirements or when low-CP and lowdigestibility crop residues are fed.

- 3. Many legumes have a deep-reaching taproot system, providing access to water and nutrients in deeper soil layers (Rao 1998; Dubeux et al. 2015), which contributes to improved cycling of both N and P (Thomas 1995; Oberson et al. 2006).
- 4. There is an enormous organismal (taxonomic) and genetic diversity in the Leguminosae (or Fabaceae) family with almost 20,000 species (Williams 1983; Lewis et al. 2005) in formerly 3, now 6 (LPWG 2017), subfamilies. This includes annuals and peren-nials, growth forms ranging from herbaceous, prostrate species (e.g. *Arachis pintoi*) to vines (e.g. *Centrosema* spp.), subshrubs (many *Stylosanthes* spp.), shrubs (e.g. *Cratylia argentea*) and trees (e.g. *Leucaena* spp.). Such diversity suggests that a very wide range of production-relevant features, in terms of adaptations to abiotic and biotic constraints, biomass production potential etc., could be expected; they warrant further exploration.
- 5. A wide range of phytochemicals (secondary metabolites) occur in many species of the Leguminosae. These are often referred to as 'antinutritional factors' when legume feeding to livestock is considered (Kumar and D'Mello 1995).

These key features imply that legumes can have a significant ecological advantage over other plant families. However, it is also via this ecological advantage that a legume can become a weed that threatens biodiversity and/or agricultural productivity and can also affect productivity via soil acidification (see below).

Tropical forage legumes and natural resources

Concern 1. Ecosystem destruction and degradation

This concern encompasses both the destruction of natural ecosystems such as forests and the degradation of areas that have already undergone land use changes, such as unproductive, mismanaged pastures. 'Prevention is better than cure' – so the initial approach to this problem should be taking measures to avoid ecosystem destruction and land degradation in the first place. Solving this issue does not require development of technology but rather application of existing appropriate land use policies and strategies.

Among them is the SI policy goal of concentrating production on existing agricultural land (<u>Garnett et al.</u> 2013; <u>The Montpellier Panel 2013</u>), thereby lowering the colonization pressure on natural ecosystems that should be considered as ecological and biodiversity reserves. Intensification, however, is usually closely linked to N

fertilization and its detrimental consequences for the environment (nitrate leaching and emission of N_2O , a potent GHG; see below).

Forage legumes can contribute to SI by providing N to the soil-plant system and high quality forage to livestock. By this, the productivity of land and livestock can be substantially increased in production systems with grasslegume pastures and/or legume-only protein banks. In Table 1 a number of examples in the tropics are presented.

There is also significant potential to increase overall land productivity via mixed-production systems such as agropastoral systems (<u>Ayarza et al. 2007</u>; <u>Boddey et al.</u> 2015), including intercropping forage legumes (<u>Hassen et al. 2017</u>), and (agro) silvopastoral systems (<u>Nair et al.</u> 2008; <u>Dubeux et al. 2015</u>). Multi-purpose legumes serve multiple roles, e.g. *Leucaena leucocephala* provides wood and forage, while *Desmodium heterocarpon* subsp. *ovalifolium* ('D. *ovalifolium*') and *Arachis pintoi* can control erosion, suppress weed growth and provide forage. <u>Dubeux et al. (2017b)</u> reviewed the role of tree legumes and their benefits in warm-climate silvopastoral systems and concluded that they were a key component for the SI of livestock systems in that climatic zone. Research has shown that, once mismanaged land has become unproductive, both herbaceous (<u>Ramesh et al. 2005</u>) and woody legumes (<u>Chaer et al. 2011</u>) can be used successfully for rehabilitation of degraded land, including degraded cattle ranching land (<u>Murgueitio et al. 2011</u>).

Concern 2. Soil degradation and loss

Soil degradation and loss are intimately linked to the previous concern, ecosystem destruction and degradation. The loss of top soil, where most soil organic carbon

Grass	Country/region	Climate/ ecosystem	Legume species	Livewei	Reference	
		5		Grass alone	With legume	-
Native (<i>Heteropogon</i> contortus)	Australia, Central Queensland	Dry subtropics	Stylosanthes humilis	83 kg/an/yr	121 kg/an/yr	Shaw and Mannetje (1970)
Native	Australia, Northern Territory	Dry tropics	Centrosema pascuorum ¹	-183 g/an/d	489 g/an/d	<u>McCown et al.</u> (1986)
Urochloa mosambicensis	Australia, Northern Queensland	Dry tropics	Leucaena leucocephala cv. Cunningham L. diversifolia	381 g/an/d ²	723 g/an/d ² 532 g/an/d ²	<u>Jones et al.</u> (1998)
Brachiaria humidicola ³	Venezuela, Táchira	Humid tropics	Desmodium ovalifolium ⁴	336 g/an/d	385 g/an/d	<u>Chacón et al.</u> (2005)
Brachiaria decumbens ⁵	Colombia, Llanos	Subhumid (savanna)	Pueraria phaseoloides	124 kg/an/yr	174 kg/an/yr	<u>Lascano and</u> Estrada (1989)
Andropogon gayanus	Colombia, Llanos	Subhumid (savanna)	Stylosanthes capitata	120 kg/an/yr 240 kg/ha/yr	180 kg/an/yr 280 kg/ha/yr	<u>CIAT (1990)</u>
Brachiaria dictyoneura ³	Colombia, Llanos	Subhumid (savanna)	<i>Centrosema</i> <i>acutifolium</i> cv. Vichada	191 g/an/d ⁶	456 g/an/d ⁶	<u>Thomas and</u> Lascano (1995)
			Stylosanthes capitata		446 g/an/d ⁶	
Brachiaria decumbens ⁵	Brazil, Mato Grosso do Sul	Subhumid (savanna)	Calopogonium mucunoides	327 kg/ha/yr	385 kg/ha/yr	<u>CNPGC (1988)</u>
<i>Pennisetum purpureum</i> cv. Kurumi	Brazil, Santa Catarina	Humid subtropical	Arachis pintoi	716 g/an/d	790 g/an/d	Crestani et al. (2013)
Brachiaria brizantha ⁷	Costa Rica, Guápiles	Humid tropics	Arachis pintoi	139 kg/an/yr ⁸ 597 kg/ha/yr ⁸	166 kg/an/yr ⁸ 736 kg/ha/yr ⁸	Hernández et al. (1995)
Brachiaria brizantha ⁷	Mexico, Veracruz	Wet-dry tropics	Cratylia argentea	580 g/an/d	839 g/an/d	González-Arcia et al. (2012)

Table 1. Effects of tropical forage legumes on liveweight gain of cattle (extracted from Rao et al. 2015).

¹Supplementation as ley during the main dry season; ²192 grazing days; ³Now classified as *Urochloa humidicola*; ⁴Now classified as *Desmodium heterocarpon* subsp. *ovalifolium*; ⁵Now classified as *Urochloa decumbens*; ⁶Means of 3 grazing cycles totalling 385 days, newly established pastures; ⁷Now classified as *Urochloa brizantha*; ⁸Mean of 2 stocking rates (low and high).

(SOC) and plant nutrients are concentrated (<u>Lal 2010</u>), leads not only to loss of a stratum that is crucial for plant production but also to oxidation of SOC and subsequent liberation of the GHG, CO₂. Since this carbon stems from recent (= not fossil) photosynthesis, it does not alter the longer-term CO₂ balance in the atmosphere. However, it is lost from a key carbon sink: soil organic matter (SOM).

Among the multiple possibilities (most of which are based on legume N contribution, soil-covering growth habit and deep root system) to contribute to the mitigation of this environmental problem, are:

- Soil conservation by: cover legumes such as *Alysicarpus vaginalis*, *Arachis pintoi* and *Desmodium 'ovalifolium'* which prevent erosion; contour-hedges with shrub species such as *D. cinereum* and *Flemingia macrophylla*; and leguminous trees such as *Erythrina* spp. and *Leucaena* spp.
- Rehabilitation of degraded soils by pioneering legumes such as *Stylosanthes* spp., *Macrotyloma axillare* and *Flemingia* spp., which are deep-rooted and adapted to infertile soils, with soil improvement resulting from cycling of minerals from deeper soil layers and enhanced concentration of SOM through litter production (Amézquita et al. 2004; Boddey et al. 2015). In the case of tannin-rich species, such as *F. macrophylla*, litter has a marked impact as it decomposes slowly (Budelman 1988) and provides a longer-lasting soil cover and slow nutrient release.
- Exploring and exploiting the potential of legumes to ameliorate compacted soil, as shown by e.g. <u>Rochester</u> et al. (2001) for *Lablab purpureus* (among other, more temperate grain legumes) and <u>Lesturguez et al. (2004)</u> for *Stylosanthes hamata*.
- Exploring and exploiting the potential adaptation of species to soil salinity. There seems to be some potential in a few genera such as *Acaciella*, *Desmanthus*, *Neptunia* and *Sesbania* (Cook et al. 2005).

Concern 3. Water degradation and loss

On a global scale, water and its decreasing availability, accessibility and quality, are major concerns (<u>Rogers et al. 2006</u>). As far as tropical pastures and forages are concerned, we see the role of legumes primarily in the following areas:

• Use of drought-adapted species, e.g. deep-rooted herbs and subshrubs such as *Centrosema brasilianum* and *Stylosanthes guianensis*; shrubs and trees such as *Cratylia argentea* and *Leucaena leucocephala* (Cook et al. 2005); or species with physiological mechanisms for avoiding and/or tolerating water stress (annual life cycle, narrow leaflets, leaf movements, tolerance of very low leaf water potentials), such as *Centrosema pascuorum* (Ludlow et al. 1983; Clements 1990).

- Reducing sedimentation of water bodies. Sedimentation is a major issue with devastating consequences in times of excessive rainfall and is, obviously, intimately linked to soil erosion by water. Consequently, the potential role of legumes consists primarily in prevention of soil erosion (see above). Additional potential lies in watershed protection through productive, N self-sufficient multipurpose trees.
- Enhancement of water infiltration via the potential amelioration effect on soil structure of legumes (see above).
- Using cover legumes to control weed growth in oil palm and rubber plantations as an attractive alternative to the use of herbicides.
- Replacing N fertilizer, at least partly, by a legume. This could reduce nitrate leaching and water eutrophication as both groundwater contamination by nitrate leaching and N-eutrophication of water bodies as a consequence of surface runoff are recognized negative consequences of N fertilization in tropical pastures (Vendramini et al. 2007).

Concern 4. Biodiversity degradation and loss

Any land use change, such as the establishment of forage species, has profound implications for biological diversity (<u>Alkemade et al. 2013</u>) in terms of plant and animal species and ecotypes, including entomofauna and the whole soil biota in the area concerned. This is particularly true if a monospecific grass sward is established, as is common in the tropics. While this is an area of considerable knowledge gaps, we claim that the inclusion of an N-fixing and, subsequently, SOM-increasing legume in a mixture with a grass will mitigate the overall negative effects of such a land-use change on biodiversity, namely entomofauna and soil biota (<u>Ayarza et al. 2007</u>). In their review which focused on temperate conditions, <u>Phelan et al. (2015</u>) reported on positive effects of legumes on the diversity and abundance of pollinating insects and earthworms.

In this context, the possible mitigating effects on biodiversity loss of using mixtures of legume species should be explored. Mixtures of herbaceous cover legumes are commonly used for weed control in Southeast Asian tree plantations, e.g. *Calopogonium* mucunoides, C. caeruleum, Centrosema pubescens (now classified as C. molle), Desmodium ovalifolium (now classified as Desmodium heterocarpon subsp. ovalifolium) and Pueraria phaseoloides (Jalani et al. 1998). Such mixtures might also improve functional biodiversity.

A related area is the role that forage legumes can play in combating agricultural pests through exudation of chemical compounds. A significant example is the increasing use of *Desmodium intortum* and *D. uncinatum* as intercrops to control maize stemborer and *Striga* spp. in the so-called push-pull systems in East Africa (<u>Khan et</u> <u>al. 2010; icipe 2015</u>).

Negative aspects of tropical forage legumes

Two negative aspects of tropical forage legumes must be recognized:

Weed potential. The danger that an exotic legume could become a serious invasive weed that threatens local biodiversity and/or affects crop production must be considered. According to available literature, this risk seems to be a particular concern in Australia, even to the point that Low (1997) suggested that introduction of exotic forage germplasm should cease with the focus changing to developing cultivars from native species. Among the factors contributing to the weed potential are (Driscoll et al. 2014): region- or production systemspecific lack of grazing or browsing animals; unpalatability or low palatability to livestock, due to presence of secondary metabolites; prolific seeding; and presence of thorns and spines. Tropical legume species currently listed among the 32 land plant species of "100 of the world's worst invasive alien species" (Lowe et al. 2004) include: Acacia mearnsii, Leucaena leucocephala, Mimosa pigra, Prosopis glandulosa and Pueraria montana var. lobata. It is well recognized that attributes which make a legume a useful pasture species are the same as those which allow it to become potentially a serious weed.

Even if a legume might not represent a risk to biodiversity on a larger scale, at the pasture level soil N accumulation following eventual legume dominance could lead to changes in species composition: nitrophilous weeds can become an agroecological problem (McIvor et al. 1996).

Soil acidification. Continuous use of legume-only or legume-dominated swards can result in soil acidification as <u>Noble et al. (1997)</u> and <u>Liu et al. (1999)</u> reported for *Stylosanthes* species in Australia and China, respectively.

It has been suggested that increased presence of a grass reduces the problem (<u>Scott et al. 2000</u>).

Tropical forage legumes and climate change

Increase in GHG emissions is claimed to be the main causal agent of climate change (Adger and Brown 1994). In low-income countries, that is, in the developing world, agriculture and land use changes are estimated to contribute 20 and 50%, respectively, to overall GHG emissions (The World Bank 2010). Climate change is expected to: (1) raise temperatures across the planet; and (2) disturb rainfall patterns, but regional differences will occur, resulting in increases of both drought-stricken and waterlogged areas, and salinization of agricultural soils (IPCC 2014; Zabel et al. 2014; Brown et al. 2015).

General strategies to cope with climate change are: adaptation to the modified climatic conditions; and mitigating GHG emissions that lead to climate change. Both are examined in relation to tropical forage legumes as follows:

Adaptation potential

We suggest that research make use of the large organismal (= taxonomic) and genetic diversity of tropical forage legumes that is available in the world's major germplasm collections, e.g. particularly those held by the Australian Pastures Genebank, CIAT (Centro Internacional de Agricultura Tropical), Embrapa (Empresa Brasileira de Pesquisa Agropecuária) and ILRI (International Livestock Research Institute). Collections can be screened for adaptation to constraints such as high temperatures and tolerance of drought, waterlogging or soil salinity (Baron and Bélanger 2007). As a result of phenotypic evaluation within the naturally available diversity, promising germplasm can be developed further via selection or breeding (Araújo et al. 2015).

In this context, existing legume germplasm collections need to be complemented by further gathering of wild germplasm in the field. Collecting missions should focus on areas which experience drought or waterlogging or soil salinity problems, i.e. areas where naturally occurring plants can be expected to have the desired adaptations for survival and productivity.

Mitigation potential

While a recent overview (<u>Peters et al. 2013</u>) concluded that tropical pastures and forages in general have the potential to play a significant role in mitigation of climate change, the following discussion refers specifically to the contribution of forage legumes.

Carbon dioxide (CO₂). The work of Fisher et al. (1994) in the Colombian Llanos showed that sown, deep-rooted tropical grasses can accumulate more SOC than native savanna, in fact, almost as much as under forest. When a legume was mixed with the grass, the amount of C stored in the soil (0-80 cm) increased by 20% to a total of 268 t C/ha. Tarré et al. (2001) reported that, in the humid tropics of Bahia, Brazil, soil C accumulation (0-100 cm soil depth) in a Brachiaria humidicola (now accepted as Urochloa humidicola)-Desmodium ovalifolium (now accepted as Desmodium heterocarpon subsp. ovalifolium) pasture over a 9-yr period was almost twice that of a B. humidicola pasture (1.17 vs. 0.66 t C/ha/yr). Contributions by nontropical permanent pastures and perennial legumes to increased C accumulation in the soil are cited in the review of Jensen et al. (2012). According to these authors, the organic N provided by the legumes fosters C accumulation. As Smith et al. (2008) and Chaer et al. (2011) showed, trees in agroforestry systems, particularly leguminous trees, have the potential to increase C accumulation in the soil considerably, as well as accumulating C in their own biomass, especially on degraded land.

On the other hand, respiration by legume roots during the energy-consuming SNF process releases substantial amounts of CO_2 to the atmosphere, even more CO_2 per unit N than is emitted during the production of industrial N fertilizer (Jensen et al. 2012). As these authors point out, however, in contrast to CO_2 from fertilizer production, CO_2 produced during SNF stems from photosynthesis, so the atmospheric CO_2 -concentration balance is not altered.

The particular role of SOM merits further emphasis. This is the most important carbon sink and can be larger than the above-ground C in a tropical rainforest (Lal 2010). If soil erodes, this eventually leads to oxidation of C to CO₂, which is released to the atmosphere (Olson et al. 2016). Therefore, perennial plants, e.g. grasses and legumes, which provide soil cover and prevent erosion, play a particularly significant role in mitigating CO₂ emissions in tropical production systems. To guarantee this environmental benefit, vegetation/pasture management must be such that there is always adequate soil stoloniferous species such cover. Creeping, as Desmodium 'ovalifolium' and Arachis pintoi that provide a dense soil cover - while supplying N-rich litter - appear to be of particular interest. It must, however, be mentioned that, because of the low C:N ratio of legumes, SOM under legume-only vegetation is less stable than under a grass-legume mixture (<u>Sant-Anna et al. 2017</u>).

Methane (*CH*₄). Methane has 25 times greater global warming potential per unit mass (100-yr time horizon) than CO₂. In agriculture, it is generated mainly by enteric fermentation, manure management and rice cultivation. By nature ruminants produce enteric CH₄ (Broucek 2014) and research is underway to determine how this might be modified. Options are either to increase the amount of meat or milk produced per unit of CH₄ emitted or to decrease the amount of CH₄ emitted per unit of feed intake through: (1) providing high quality forage, mainly in terms of CP concentration and digestibility; and (2) improving livestock breeds that are able to respond to improved forage quality with increased productivity (Gerber et al. 2013).

In a recent meta-analysis, Lee et al. (2017) showed that rising temperatures lead to decreased nutritive value of grasses and increased CH₄ emissions by ruminant livestock, which worsens the global warming scenario. On the other hand, forage legumes have high nutritive value and can contribute to lower emissions of CH₄ per unit of livestock product or unit of feed ingested. A study by <u>Molina et al. (2016)</u> of methane emissions of Lucerna heifers fed a *Leucaena leucocephala*-stargrass mixture or grass only demonstrated the benefits of the legume in the diet in reducing methane emissions per unit gain. The optimal situation is to have improved livestock feeding, based on high quality forage including legumes, combined with improved livestock breeds that can more efficiently use such improved feed.

In addition to this general quality-based role of forage legumes regarding enteric CH₄, another meta-analysis (Jayanegara et al. 2012) showed that polyphenols such as condensed tannins, i.e. secondary metabolites that occur in many tropical forage legumes, decrease CH₄ emissions. According to an analysis based on 22 in vivo studies, ruminants fed warm-climate legumes produced less CH₄ per kg OM intake than ruminants fed cold-climate legumes, C3 grasses and C4 grasses (Archimède et al. 2011). Low-molecular weight tannins, such as those in L. leucocephala (Molina et al. 2016), can also play a role. It is important to ensure that tannins in the diet do not reduce protein digestibility, compromising animal intake and thus its performance, which in turn will affect CH₄ emissions per unit of livestock product. Working with subterranean clover (Trifolium subterraneum) Kaur et al. (2017) showed that a plant breeding approach to reduce methanogenesis has potential.

Nitrous oxide (N_2O). Nitrous oxide has 300 times greater global warming potential per unit mass (100-yr time horizon) than CO₂. Its production by soil microorganisms during nitrification and denitrification processes is very much related to the use of N fertilizers in agriculture (Subbarao et al. 2013). In their meta-analysis, Jensen et al. (2012) concluded that there is a tendency for lower N₂O production from soil under legumes than from systems based on industrial N fertilizer, depending on the amount of N fertilizer applied. This seems to be an area of considerable knowledge gaps in relation to tropical forage legumes.

In view of the recent detection of biological nitrification inhibition (BNI) in some tropical forage grasses, particularly *Brachiaria* (now *Urochloa*) *humidicola* (Subbarao et al. 2009; 2017), the challenge is to determine whether such a mechanism might also exist in tropical forage legumes. It might then be possible to exploit the synergy between SNF and BNI to the benefit of both agriculture and the environment. Due to BNI, symbiotically fixed N might be available for longer periods and less prone to loss by nitrate leaching and N₂O production.

Discussion and Conclusions

Ecosystem services

In the preceding sections, we showed that tropical forage legumes have considerable potential to increase productivity of forage-based livestock systems, while providing benefits to the environment. The environmental benefits, subsumed under 'ecosystem services', comprise positive effects on: soil conservation and soil chemical, physical and biological properties; water balance; mitigation of global warming and of groundwater contamination; saving of fossil energy; functional biodiversity (soil, entomofauna); and rehabilitation of degraded land. The combination of these features makes tropical forage legumes particularly valuable at all levels of the system because of their interaction with plants, soil, animals and the atmosphere. This environmental role could be considered as a 'new' important dimension of tropical forage legumes.

A crucial aspect, however, is: During past decades the beneficial role of tropical forage legumes was promoted with the sole focus on livestock production and soil fertility; what must be done to have legume-based technologies more readily adopted by farmers now that general environmental benefits are recognized? *Legume technology adoption and payment for ecosystem services*

In their review paper, which examined the role of forage legumes in general (though they focused primarily on temperate zones), Phelan et al. (2015) reported a low and even declining use of forage legumes. We must recognize that in the tropics adoption of legume-based technologies has, in general, been disappointing - in spite of many success stories with tropical forage legumes worldwide (see the 33 contributions in Tropical Grasslands Vol. 39, No. 4, 2005; goo.gl/Qf5VJu). The reasons were analyzed by Shelton et al. (2005) and include a number of issues that should be taken into account when planning R&D programs promoting the use of tropical forage legumes. A particularly important issue is the organization of efficient seed production systems. The lack of seed availability is often cited as a key reason for adoption failure and the resulting vicious circle (lack of robust demand - lack of interest of the private seed production sector – lack of seed production and availability - lack of adoption) needs to be broken. Successful results have been achieved with contracting farmers for forage legume seed production and farmer to farmer seed sales, e.g. in Thailand, India and Bolivia. For large-scale adoption it will be essential to develop systems which ensure high seed quality and are commercially viable (Shelton et al. 2005).

We doubt that an eventual recognition of the 'new' ecosystem services role of legumes will modify farmers' lack of enthusiasm for legumes to a marked extent. Although promotional and educational activities, along with results from further research involving farmer participation, might be helpful, we expect that constraints imposed by the need for management skills and investments will remain, unless attractive economic incentives are offered to farmers (White et al. 2013). Such incentives should not be restricted to legume-based technologies but should extend to all tropical forage technologies which provide environmental services. We suggest that schemes of payment for ecosystem services (PES) (Pagiola et al. 2004; Van Noordwijk and Leimona 2010), applicable to both smallholders and large livestock producers, be explored, developed and implemented.

The need for life cycle assessments

Inputs of N are necessary in all pastures if livestock productivity is to be increased, such as within the concept of SI. Basically, there are 2 options: (1) planting legumes with SNF capability in mixtures with grasses; and (2) applying industrial N fertilizers to grass-only swards.

Greenhouse gas emissions from both approaches should be measured. We suggest that full life cycle assessments for tropical pastures addressing the whole carbon footprint (Eshel et al. 2014) should be performed. In their temperate climate-focused review, <u>Phelan et al. (2015)</u> reported that CO₂-equivalent emissions for *Trifolium repens*-grass pastures were 11-23% lower than for Nfertilized grass. Such life cycle assessments must include the need for fossil energy and any benefits to any subsequent crop in a rotational system (<u>de Vries and de Boer 2010</u>; Jensen et al. 2012).

Research needs

The suboptimal adoption of forage legume technologies in the past – when only forage dry matter and/or livestock production was considered – has led to a substantial decrease in research on tropical forage legumes during the last 2 decades. We argue that, in view of current environmental concerns, this research should be resumed with adequate funding support at national and international levels.

We have shown that a substantial body of evidence suggests that forage legumes have potential to contribute significantly to environment-friendly agricultural land use and sustainably intensified livestock production in the tropics. However, there is still a lack of hard data, and several statements in our analysis are not yet well substantiated and need to be verified and confirmed. Further research is required to provide decision makers with a solid database on the ecosystem services from utilization of tropical forage legumes. Priorities in different regions will depend on differences in climate, soil types, land use, production systems etc. Preferably, such research should be conducted within a coordinated network or consortium, e.g. similar to those European initiatives with focus on temperate legumes (Lüscher et al. 2014).

We have compiled the following list of 'research for development' themes on tropical forage legumes as a result of our analysis:

- life cycle assessments to compare the carbon footprints of livestock feeding based on forage legumes with that based on N fertilizer in different production systems;
- potential of legumes for enhancing functional biodiversity, including in multi-species mixtures;
- further understanding of the potential of forage legumes in (1) crop-livestock systems, (2) soil stabilization and (3) reversing land degradation;
- further understanding of the impact of legumes on associated vegetation (species composition);

- assessment of the impact of promising legume species on rumen methanogenesis;
- identification of tanniniferous legumes which concurrently provide high quality forage in terms of digestibility in the rumen and reduced methane emission intensity;
- identification of anti-methanogenic compounds other than tannins in legume forage;
- assessment of the BNI potential of forage legumes;
- development of methodologies for payment for ecosystem services;
- optimization of SNF via enhanced exploration and exploitation of rhizobia diversity; and
- targeted collection of wild legume germplasm for development of varieties with improved adaptation to climate variability and change.

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Research Paper

Soil attributes of a silvopastoral system in Pernambuco Forest Zone *Atributos del suelo en un sistema silvopastoril en la "Zona da Mata", Pernambuco, Brasil*

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Abstract

This research evaluated soil properties in a silvopastoral system using double rows of tree legumes. Treatments were signalgrass (*Brachiaria decumbens*) in monoculture or in consortium with sabiá (*Mimosa caesalpiniifolia*) or gliricidia (*Gliricidia sepium*). Treatments were arranged in a complete randomized block design, with 4 replications. Response variables included chemical characteristics and physical attributes of the soil. Silvopastoral systems had greater (P<0.001) soil exchangeable Ca (gliricidia = 3.2 and sabiá = 3.0 mmolc/dm³) than signalgrass monoculture (2.0 mmolc/dm³). Water infiltration rate was greater within the tree legume double rows (366 mm/h) than in signalgrass (162 mm/h) (P = 0.02). However, soil moisture was greater in signalgrass pastures (15.9%) (P = 0.0020) than in silvopastures (14.9 and 14.8%), where soil moisture levels increased as distance from the tree rows increased. Conversely, the light fraction of soil organic matter was greater within the tree legume double rows than in the grassed area (P = 0.0019). Long-term studies are needed to determine if these benefits accumulate further and the productivity benefits which result.

Keywords: Fertility, legumes, soil physics, trees.

Resumen

Entre enero 2012 y diciembre 2013 en Itambé, Pernambuco, Brasil, se evaluaron algunas propiedades físicas y químicas del suelo en un sistema silvopastoril, utilizando filas dobles de leguminosas arbóreas. Los tratamientos consistieron en *Brachiaria decumbens* sola o en asociación con sabiá (*Mimosa caesalpiniifolia*) o gliricidia (*Gliricidia sepium*) en un diseño de bloques completos al azar, con 4 repeticiones. Los sistemas silvopastoriles presentaron mayor contenido (P<0.001) de calcio intercambiable (gliricidia = 3.2 y sabiá = 3.0 mmolc/dm³) comparados con la gramínea sola (2.0 mmolc/dm³). La tasa de infiltración de agua fue mayor en el suelo dentro de las filas dobles de los árboles leguminosos (366 mm/h) en comparación con la gramínea sola (162 mm/h) (P = 0.02). No obstante, la humedad fue más alta en el suelo con gramínea (15.9%) (P = 0.0020) comparada con los sistemas silvopastoriles (14.9 y 14.8%, respectivamente para sabiá y gliricidia). La humedad en el suelo aumentó con la distancia a partir de la línea de árboles. Por el contrario, la fracción ligera de la materia orgánica del suelo fue mayor (P = 0.0019) dentro de las filas dobles de árboles (0.071 mg/kg) comparada con el suelo fuera de la línea de árboles. Se requieren estudios a largo plazo para determinar si estos beneficios continuan acumulándose y si resultan en mayor productividad.

Palabras clave: Árboles, fertilidad, física del suelo, leguminosas.

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Introduction

Good soil physical characteristics are essential to ensure satisfactory crop and pasture productivity. Pasture soils tend to have greater soil density than preserved vegetation soil, presumably due to trampling by animals (Vitorino 1986), which can also have an impact on the water infiltration rate and soil moisture holding capacity, both of which have significant effects on pasture productivity. The amount of water that infiltrates and flows over the ground is directly related to soil physical properties such as density, and the existing vegetative cover (Lanzanova et al. 2007).

Soil organic matter (SOM) has a major influence on ecosystem productivity because it affects chemical and physical characteristics of soils. Since SOM is the net result of soil processes occurring in the long term, it is difficult to detect early changes if analyzing total SOM (<u>Haggerty and Gorelick 1998</u>). The light fraction of the SOM is formed by plant and animal residues in the early stages of decomposition. It represents recent changes in land management and can detect early changes in SOM dynamics (<u>Jinbo et al. 2007</u>; <u>Rangel and Silva 2007</u>). Increases in ecosystem primary productivity lead to increasing residue deposition, both above- and belowground.

Silvopastoral systems improve soil physical attributes such as soil aggregates, soil density and water infiltration rates (Carvalho et al. 2004). Litter deposition from tree foliage is a major pathway for recycling of nutrients in a silvopastoral system (Apolinário et al. 2016). Limited nitrogen (N) availability in warm-climate grasslands is one of the major limiting factors to increases in productivity (Vendramini et al. 2014), and N addition via litter represents a significant input and might result in greater ecosystem primary productivity. Tree legumes such as sabiá (Mimosa caesalpiniifolia Benth.) and gliricidia [Gliricidia sepium (Jacq.) Kunth] can be used in silvopastoral systems (Souza and Espíndola 2000; Vieira et al. 2005; Apolinário et al. 2016; Costa et al. 2016). Besides biological N2 fixation, litter deposition and decomposition are important sources of nutrients to be reused by the system (Apolinário et al. 2016).

Tree legumes can provide extra alternative income through the sale of fencing posts and firewood (Apolinário et al. 2015). Incorporating tree legumes in silvopastoral systems can also provide other ecosystem services including the maintenance of biodiversity, improvement of water and nutrient flow, enhancement of soil quality, reduction of soil erosion, improvement of C storage and provision of green areas for urban society (Kemp and Michalk 2005). Given the economic and environmental importance of these systems, this study aimed to evaluate the chemical composition and physical properties of soils in signalgrass pastures [*Brachiaria decumbens* Stapf; now: *Urochloa decumbens* (Stapf) R.D. Webster], in association with tree legumes in the coastal region ("Zona da Mata") of Pernambuco State, Brazil.

Materials and Methods

The research was conducted at the Experimental Station of the Agronomic Institute of Pernambuco (IPA), located in Itambé, Pernambuco, Brazil. Average annual rainfall is 1,300 mm, and average annual temperature is 25 °C (CPRH 2003). The climate is sub-humid, the topography is undulating and the soil of the study area is classified as Ultisol (red-yellow dystrophic Argissol according to the Brazilian Soil Classification or Paleudult or Ferric Luvisol according to FAO World Reference Base) (Jacomine et al. 1972; Embrapa 2006). Initial soil chemical characteristics of the experimental area were: pH in water (1:2.5) 5.5; P (Mehlich-I) 2.2 mg/dm³; K 1.3 mmolc/dm³; Ca 27 mmolc/dm³; Mg 20 mmolc/dm³; Na 1.4 mmolc/dm³; Al 2.7 mmolc/dm³; H+Al 61.7 mmolc/ dm³; and SOM 44.2 g/kg. Average monthly rainfall for the experimental years is shown in Figure 1.

Three treatments were tested in a complete randomized block design with 4 replications. Treatments included: 1) sabiá with signalgrass; 2) gliricidia with signalgrass; and 3) signalgrass monoculture. Each experimental unit measured 660 m² (33 x 20 m). Tree legumes (sabiá and gliricidia) were established in 2008 in double rows spaced at 10.0 m (between double rows) x 1.0 m (between rows) x 0.5 m (within rows). Each plot contained 3 double rows. The signal grass was growing throughout the area of each plot, but reduced growth occurred between the individual tree legume rows that formed the double rows ("within tree legume double rows" from here on), especially under sabiá trees. Livestock were introduced to the paddocks when the sward height reached 60 cm, and remained until the stubble height of the grass was reduced to 10-15 cm.

Soils from tree legume paddocks were sampled in September 2012 in order to determine the chemical composition. Samples were collected in 2 transect lines perpendicular to the tree rows. Along each transect, 5 different points were sampled (0, 1, 2, 3 and 4 m distance from each tree double row) giving 30 samples per plot (Figure 2). Paddocks with signalgrass monoculture were sampled randomly at 5 sites. All soil samples were taken from the 0–20 cm soil layer. Soil samples to determine bulk density and soil gravimetric moisture were collected

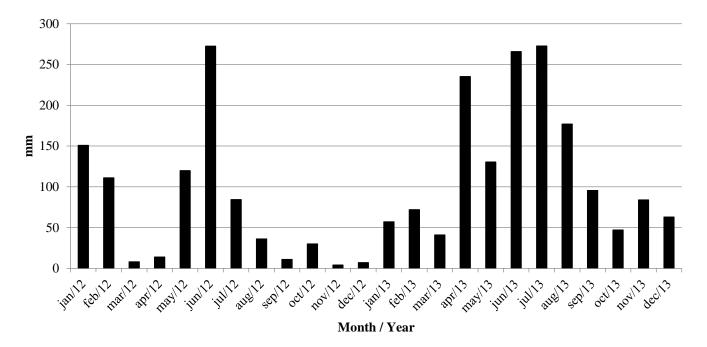
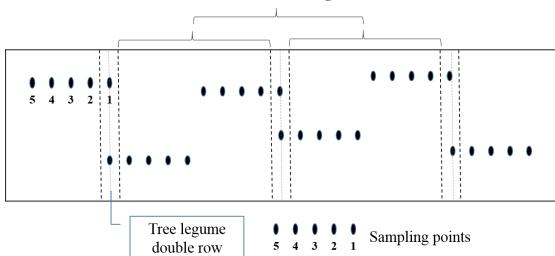


Figure 1. Rainfall (monthly averages) in the experimental area during the research period. Source: Meteorological data collected at the experimental site.

in May 2013, using the same sampling protocol (perpendicular transects) described to collect the soil fertility samples, and the same soil depth. Undisturbed soil cores were collected using volumetric rings. Samples were dried in an oven at 105 °C for 24 hours, following methodology described by Embrapa (1979).

Light fraction of SOM was determined by weighing 50 g of soil (samples collected for fertility analyses), which

were macerated and sieved through a 0.5 mm sieve, and then put into a 0.053 mm sieve and washed in running water. The retained material was then transferred to containers filled with water, where it remained undisturbed for 24 h for density separation (heavy and light fraction). The supernatant (floating) material was retrieved in 0.053 mm mesh, dried at 65 °C for 72 h, and weighed on a precision scale (Correia et al. 2015).



Grassed area between legume rows

Figure 2. Location of soil sampling points relative to tree legume rows in the silvopasture treatments.

Water infiltration rate (WIR) was determined in January 2013. Infiltrometers made of concentric rings (Bouwer 1986) were placed at 2 specific points in each tree legume paddock: 1) within tree legume double rows (sampling point 1); 2) in the middle of the grassed area (sampling point 5). A total of 48 samples were collected (2 replicates for each sample location within each silvopastoral system, and 4 samples for each signal-grass plot). Water infiltration rate was determined when the rate was constant, using the following equation:

WIR (mm/min) = L2 - L1 (mm) / time (min)

where: L2 is water at the beginning of the measurement and L1 is the remaining water in the tube after the time spent measuring.

Soil attributes were analyzed using PROC MIXED (SAS 2007). A complete randomized block design was used to compare signalgrass monoculture with the silvopastoral systems. When transects were analyzed, the transect points were considered split-plot and the main plot the vegetation cover, with both being fixed effects. In all analyses, blocks were considered a random effect. Significance was declared at 5% probability. LSMEANS were compared using the PDIFF procedure and adjusted Tukey test.

Results

Soil fertility

While soil chemical composition was affected by vegetation cover (Table 1), levels of most nutrients were similar in all treatments (P>0.05). Soil pH was greater in the signalgrass monoculture than in the 2 grass-legume tree pastures. Soil exchangeable Ca was greater in the grass-legume tree pastures than in the grass-only pasture,

and soil exchangeable Na was greater in the signalgrasssabiá pasture than in the other pastures (Table 1). When com-paring the sampling points in relation to the distance from the rows of legumes, there was no significant effect for the response variables evaluated, except for pH (Figure 3), where values increased exponentially as distance from the legume rows increased, with a peak at 3 m.

Water infiltration rate

Water infiltration rate was higher within the tree legume double rows of gliricidia and sabiá (356 and 366 mm/h, respectively; Figure 4) than in the signalgrass monoculture (162 mm/h) and in the grassed area of the signalgrass-sabiá (128 mm/h) treatment.

Gravimetric moisture

Soil moisture (Table 2) levels were higher (P<0.05) in the signal grass monoculture than in the mixed pastures; in the mixed pastures soil moisture increased as distance from the tree rows increased (P<0.05; Table 3).

Soil density was not affected by type of pasture (P = 0.58) (Table 2), but in the mixed pastures soil density increased as distance from the tree rows increased (P<0.05; Table 3).

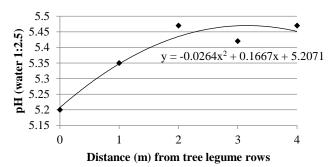
Light fraction of soil organic matter

Light fraction of SOM was unaffected by pasture type (P = 0.22), but within the silvopastoral treatments, light fraction of SOM was greater in the sabiá treatment than under gliricidia (64 vs.45 mg/kg, respectively; P = 0.002) (Table 4). The light fraction of SOM was greater under the trees than in the grass area (71 vs. 50 mg/kg, respectively; Table 3).

Table 1. Soil chemical analyses (0-20 cm layer) in signalgrass, signalgrass-gliricidia and signalgrass-sabiá pastures.

Treatment	pН	Р	K	Mg	Ca	Na	Al	H + Al	С	OM
	(water – 1:2.5)	(mg/dm ³)			(mmo	olc/dm ³)			(g/	kg)
Signalgrass	5.8 a	1.6 a	1.4 a	2.0 a	2.0 b	0.1 b	0.3 a	5.6 a	22.0 a	48.5 a
Gliricidia	5.4 b	2.5 a	1.7 a	2.0 a	3.2 a	0.1 b	0.3 a	6.5 a	29.3 a	43.4 a
Sabiá	5.4 b	2.5 a	1.7 a	1.9 a	3.0 a	0.3 a	0.3 a	6.4 a	23.6 a	40.7 a
Probability	0.02	0.26	0.17	0.87	0.001	0.02	0.91	0.15	0.23	0.71
CV (%)	3	38	85	14	12	67	51	10	23	30

Values followed by the same letter within columns do not differ by Duncan's test (P>0.05). OM = organic matter.



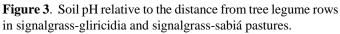


Table 2. Soil moisture and density (0–20 cm layer) in signalgrass monoculture, signalgrass-gliricidia and signalgrass-sabiá pastures.

Treatment	Moisture (%)	Density (g/cm ³)
Signalgrass	15.9 a	1.21 a
Gliricidia	14.9 b	1.22 a
Sabiá	14.8 b	1.19 a
Probability	0.002	0.74
CV (%)	3.19	4.7

Values followed by the same letter within columns do not differ by Duncan's test (P<0.05).

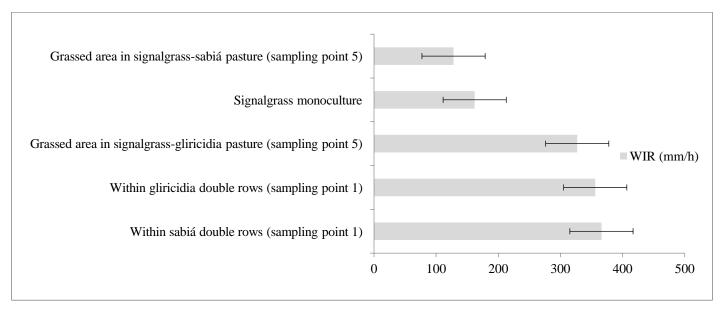


Figure 4. Water infiltration rate (mm/h) in signalgrass monoculture and grassed areas in signalgrass-gliricidia and signalgrass-sabiá pastures, and within gliricidia and sabiá double rows in the mixed pastures. The bars represent the standard error.

Table 3. Effect of distance from tree legume rows on soil moisture, soil density and soil organic matter (SOM) light fraction (0–20 cm layer) in signalgrass-gliricidia and signalgrass-sabiá pastures.

Distance (m) from tree rows	Soil moisture (%)	Soil density (g/cm ³)	SOM light fraction (g/kg)
0	14.5 b	1.18 b	0.071 a
1	14.1 b	1.19 b	0.051 b
2	15.2 ab	1.19 b	0.056 b
3	14.8 ab	1.22 ab	0.052 b
4	15.5 a	1.24 a	0.042 b
Probability	0.04	0.07	0.02
CV (%)	9.6	4.0	32.5

Values followed by the same letter within columns do not differ by Duncan's test (P>0.05).

Treatment	SOM light fraction (g/kg)
Gliricidia	0.045 b
Sabiá	0.064 a
Probability	0.002
CV (%)	32.4

Table 4. Soil organic matter (SOM) light fraction (0–20 cm layer) in signalgrass-gliricidia and signalgrass-sabiá pastures.

Values followed by the same letter within columns do not differ by Duncan's test (P>0.05).

Discussion

This study has provided some interesting results on changes in soil parameters when tree legumes are introduced into a pure grass pasture. They contribute to our knowledge of how the legumes alter the soils in conjunction with an associated grass.

Soil fertility

As in our study, <u>Carvalho et al. (2003)</u> reported increased soil exchangeable Ca in silvopastoral systems 5 years after establishment, and attributed this increase to the greater accumulation of litter produced by the trees. Similarly, <u>Camarão et al. (1990)</u> suggested that the increase in soil exchangeable Ca in silvopastoral systems might be explained by the increased above- and below-ground litter deposition. <u>Xavier et al. (2003)</u> also observed an increase in soil exchangeable Ca in signalgrass-*Acacia mangium* pasture compared with pure signalgrass.

The reduction in soil pH in the mixed pastures recorded in our study is in contrast with the findings of <u>Oliveira et</u> <u>al. (2000)</u>, <u>Andrade et al. (2002)</u>, <u>Xavier et al. (2003)</u> and <u>Dias et al. (2006)</u>, where soil pH was not affected by the introduction of trees. <u>Dias et al. (2006)</u> also studied soil chemical composition of grass-tree legume pastures in relation to the distance from the tree trunk and found variations in soil pH and levels of P, K, Ca and Mg tending to increase or decrease, depending on the legume species, planting density and biomass production.

In silvopastoral systems, most litter deposition occurs near the tree trunks (<u>Silva et al. 2013</u>), which might influence the reduction of soil pH. Greater litter accumulation leads to greater amounts of litter nutrients being mineralized. As a result, more leaching of exchangeable bases due to release of anions from OM might occur (<u>Balbinot et al. 2010</u>). However, <u>Pavan et al. (1986</u>) obtained an increase in soil pH in an area with greater litter deposition. Several studies on silvopastoral systems indicated that the benefits brought by the trees to soil fertility of the pastures tend to increase over time with development of the trees, increase in litter deposition and accumulation of animal waste, provided that the system is appropriately managed (<u>Balbino et al. 2012; Padovan and Pereira 2012; Loss et al. 2014</u>).

Water infiltration rate

Greater WIR in the signalgrass area in consortium with gliricidia might be due to the fact that this legume has a deeper root system, providing advantages such as increased water absorption and greater efficiency in the search for nutrients, resulting in its high performance as fodder for livestock (<u>Abdulrazak et al. 1997</u>; <u>Ondiek et al.</u> 1999; Juma et al. 2006). A more specific study of the root systems of gliricidia and sabiá is necessary to better understand the influence of root properties (length, depth and density) on WIR.

Silvopastoral systems allow increases in SOM because of greater litter deposition from trees, and Bell et al. (2011) indicated that greater litter deposition increases soil macroporosity, contributing to improved water infiltration and aeration. Moisture, biological activity and vegetation cover can also influence soil responses, such as the WIR (Carduro and Dorfman 1988). Dunger et al. (2005) reported that silvopastoral systems provide a favorable microclimate to increase soil microfauna, which tend to seek shaded and humid habitats. An increase of Coleoptera beetles in association with the introduction of legumes from the genus Mimosa in pastures has been reported by Dias et al. (2007). These beetles dig underground galleries in order to nest, thus providing the opportunity for greater water infiltration (Miranda et al. 1998).

Increased height and density of tree legumes in the experimental area reduced the transit of grazing cattle through the rows, which might explain the lower soil density at these points (Table 3). The WIR was greater along tree legume rows as compared with the grazed area under the effects of treading by animals, as indicated with changes in soil density. These data corroborate those of Lanzanova et al. (2007), who studied the effects of grazing on water infiltration rates in soils, finding greater WIR values in ungrazed areas and decreasing values as grazing became more intense. In our research, the increases in soil density as distance from tree legumes increased (Table 3) was reflected in decreases in WIR. Bertol et al. (2001) showed that heavy clay soils have a low percentage of the pore volume occupied by air, which leads to greater rates of runoff water, lower retention of water and consequently lower infiltration capacity. Prevedello (1996) also pointed out that the reduction in WIR with time can be influenced by factors that operate on the soil surface, such as surface sealing due to the impact of raindrops, which may be reduced by the canopy of tree legumes. Roots of tree legumes in the silvopastoral systems used in this experiment might favor soil physical aspects, maintaining and improving soil structure and increasing WIR (Hernández 1998).

Excretion of organic acids and inorganic compounds (e.g. P and K) by roots can influence soil characteristics, as they allow for increased dissolution of mineral substances and contribute to the development of rhizosphere microorganisms (Cintra et al. 1999). Roots can also favor SOM accumulation, as Lehmann and Zech (1998) found that the litter produced by the renewal of roots adds about 20–50% of the total root biomass to the SOM pool, while only 10–20% of litter arising from the aerial parts is transformed into SOM (Schroth et al. 1999). Since roots are more recalcitrant than leaves and stems, a greater proportion of original root biomass ends up in the SOM pool than leaves and stems.

Gravimetric moisture

The greater soil moisture in signalgrass monoculture was probably due to the competition by different species for water. Legumes are less efficient in water usage than C4 grasses. On average, legumes use 800 kg of water to produce 1 kg of dry matter, while C4 plants use 300 kg of water to produce the same amount of DM (<u>Taiz and</u> <u>Zeiger 2004</u>; <u>Marenco and Lopes 2009</u>). Plant species have a marked influence on water availability in silvopastoral systems and <u>Vanzela and Santos (2013</u>) highlighted that the use of eucalypts in silvopastoral systems increased competition for water and nutrients between the trees and the associated grass.

Andrade and Valentim (1999) showed that shading is a positive factor in maintaining soil moisture, resulting in satisfactory forage development in silvopastoral systems. In natural shading conditions, however, trees also compete with one another and the grass for light, water and nutrients. Therefore, the water requirements of the tree legumes might have contributed to reduced soil moisture near the trees in the current research.

Another aspect that should be highlighted is the fact that, during the collection period, the grass monoculture was approximately 60 cm tall, which provided 100% ground cover, helping to maintain soil moisture. In the silvopastoral systems, tall trees with dense canopies might have compromised production of signalgrass, which has only moderate shade tolerance and might suffer production loss due to shading (Schreiner 1987). In contrast to this, <u>Aguiar et al. (2006)</u> recorded greater soil moisture in silvopastoral systems compared with agrosilvopasture (combination of trees, crops and livestock, grown on a particular site) and intensive cultivation. <u>Perin et al. (2000)</u> also observed greater soil moisture when soil was covered with a thick litter layer of herbaceous legumes.

The increase in soil moisture as distance from the tree legume rows increased meant that grasses growing in the middle of the grass strips suffered reduced competition for soil moisture from the trees, while still having some shade to assist retention of soil moisture (Table 3). Near tree rows, there was reduced soil cover because of greater competition for resources between herbaceous and woody vegetation.

Soil density

Average soil density was 1.2 g/cm³, which is adequate for root development (<u>Alvarenga et al. 1996</u>; <u>Corsini and</u> <u>Ferraudo 1999</u>). According to <u>Argenton et al. (2005</u>), characterization of soil density depends on its textural class and <u>Rosenberg (1964</u>) and <u>Cintra and Mielniczuk</u> (<u>1983</u>) suggest that each soil type has a critical density, which can reduce or even prevent root development. <u>Reichert et al. (2003</u>) showed that 1.4 g/cm³ is considered the critical soil density for satisfactory growth of the root system of plants in clay soils, but <u>Reinert et al. (2008</u>) indicated a greater soil density (1.85 g/cm³) as critical for legumes and other vegetables in clayey soils.

The lower soil density near the trees (Table 3) can be attributed to the existence of microfauna near the trees (Miranda et al. 1998; Dunger et al. 2005; Dias et al. 2007) as well as a greater SOM accumulation between trees, increasing the amount of soil aggregates. Iori et al. (2012) studied soil density and soil moisture in degraded pastures, banana cultivation, a silvopastoral system and preserved forest. They found greater soil moisture in less dense soil, which can be correlated with the shading potential and greater SOM in these areas. Beltrame et al. (1981) stated that soil moisture affects the cohesion between soil particles, with increases in aggregation when soil moisture is limited, which hinders their separation by external forces (Silveira et al. 2010).

Light fraction of soil organic matter

While vegetation cover did not affect the light fraction of SOM (P = 0.22), in the mixed pastures sabiá presented greater values of SOM than gliricidia (Table 4). <u>Chan et al. (2002)</u> and <u>Zinn et al. (2005)</u> observed that SOM stocks are directly related to residue inputs, their rate of decomposition and SOM fractionation. They pointed out that the replacement of conventional farming systems

with improved systems, such as silvopastures, changes the dynamics of litter accumulation and litter decomposition rate, and consequently generates greater increases in the light fraction of SOM. Similarly, <u>Maia et</u> <u>al. (2008)</u> showed greater amounts of light fraction of SOM in silvopastoral systems (38.2 g/dm³) than in conventional tillage (28.4 g/dm³), because of greater litter input from trees. The amount of light fraction in the system is directly related to the litter deposited on the soil. Light fraction of SOM is composed of litter and organic matter in intermediate stages of decomposition (<u>Souza et</u> <u>al. 2006</u>) and its level at any given time is the net balance between its deposition and decomposition (<u>Fraga 2002</u>).

Phenolic substances found in plants often influence litter decomposition rate and, consequently, nutrient cycling, affecting the composition and activity of decomposing communities of the system (<u>Hättenschwiler</u> and <u>Vitousek 2000</u>). Among these substances, flavonoids are characterized by their recalcitrance, with condensed tannin (CT) concentration usually correlating with low decomposition rates (<u>Burhenne et al. 2013</u>). <u>Nozella</u> (2001) found high levels of condensed tannins (near 6.9 g/kg DM in gliricidia), while <u>Balogun et al. (1998</u>) determined mean values of 0.8%. <u>Beelen (2002</u>), however, showed greater values in sabiá, reaching up to 20.1%. Greater CT concentration in sabiá might explain the greater light fraction of SOM observed in the silvopasture with this species, compared with the one with gliricidia.

Conclusions

This study has shown that incorporation of tree legumes in rows within a signalgrass pasture can improve soil chemical composition over time as well as increasing WIR in the soil, and the concentration of light fraction SOM near the trees. These findings indicate that silvopastoral systems using tree legumes can potentially serve as greater C sinks than pure grass pastures as well as providing other services to farmers. However, long-term results coupled with life cycle assessments are necessary to determine what productivity increases will result.

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Research Paper

Germination of tropical forage seeds stored for six years in ambient and controlled temperature and humidity conditions in Thailand

Germinación de semilla de forrajeras tropicales durante seis años de almacenamiento bajo condiciones ambientales y condiciones de temperatura y humedad controladas en Tailandia

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Abstract

The germination performances of fresh seed lots were determined for 5 tropical forage species: Mulato II hybrid brachiaria [Urochloa ruziziensis (syn. Brachiaria ruziziensis) x U. decumbens (syn. B. decumbens) x U. brizantha (syn. B. brizantha], Mombasa guinea [Megathyrsus maximus (syn. Panicum maximum)], Tanzania guinea [M. maximus (syn. P. maximum)], Ubon paspalum (Paspalum atratum) and Ubon stylo (Stylosanthes guianensis), stored under ambient conditions in Thailand (mean monthly temperatures 23-34 °C; mean monthly relative humidity 40-92%) or in a cool room (18–20 °C and 50% relative humidity) for up to 6 years. The first paper of this study showed all seeds, except unscarified Ubon stylo seed, were dead after a single year of storage in ambient conditions. This second paper shows that cool-room storage extended seed viability, but performance varied considerably between species. Germination percentage under laboratory conditions declined to below 50%, after 3 years storage for Mombasa guinea seed and Tanzania guinea seed, 4 years for Ubon paspalum seed and 4–5 years for Mulato II seed. Ubon stylo seed maintained high germination for 5 years, in both cool-room storage (96%) and ambient-room storage (84%). Apparent embryo dormancy in acid-scarified Mulato II seed steadily increased with time in cool-storage and this seed had to be acidscarified again each year at the time of germination testing to overcome dormancy. Physical dormancy of Mulato II seeds, imposed by the tightly bound lemma and palea in unscarified seed, was not overcome by length of time in coolstorage and these seeds had to be acid-scarified to induce germination. Hardseeded percentage in Ubon stylo seed remained high throughout the study and could be overcome only by acid-scarification. The difficulties of maintaining acceptable seed germination percentages when storing forage seeds in the humid tropics are discussed.

Keywords: Embryo dormancy, hardseededness, humid tropics, seed storage, seed viability.

Resumen

En Tailandia se determinó la germinación de semilla de 5 cultivares de forrajeras tropicales: *Urochloa* híbrido cv. Mulato II, *Megathyrsus maximus* cv. Mombasa, *M. maximus* cv. Tanzania, *Paspalum atratum* cv. Ubon, y *Stylosanthes guianensis* cv. Ubon stylo, almacenadas bajo condiciones ambientales (temperaturas promedio mensuales 23–34 °C; humedad relativa 40–92%) o controladas en cuarto frío (18–20 °C; 50% humedad relativa) durante 6 años. Mientras en un estudio previo se encontró que bajo condiciones ambientales todas las semillas, excepto las de Ubon stylo no escarificadas con ácido, perdieron su viabilidad después de 1 año de almacenamiento, en este segundo estudio se encontró que el almacenamiento en cuarto frío prolongó su viabilidad, aunque con una alta variabilidad entre especies. La germinación bajó a <50% después de 3 años de almacenamiento para *M. maximus* cvs. Tanzania y Mombasa, 4 años

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para *Paspalum atratum* cv. Ubon y 4–5 años para *Urochloa* híbrido cv. Mulato II. La semilla de *S. guianensis* cv. Ubonstylo mantuvo una alta germinación durante 5 años de almacenamiento tanto en cuarto frío (96%) como bajo condiciones ambientales (85%). La dormancia del embrión en las semillas de cv. Mulato II, escarificadas con ácido, aumentó constantemente con el tiempo de almacenamiento en cuarto frío; por tanto, para romperla fue necesario escarificar la semilla con ácido nuevamente cada año en el momento de la prueba de germinación. De la misma forma, la dormancia física de las semillas del cv. Mulato II impuesta por la lemma y pálea fuertemente unidas en semillas no escarificadas con ácido, no se rompió con el tiempo de almacenamiento en cuarto frío, por lo que fue necesario escarificar con ácido para inducir la germinación. El porcentaje de semilla dura de *S. guianensis* cv. Ubon-stylo permaneció muy alto durante todo el estudio y la germinación solo se pudo inducir mediante escarificación con ácido. Se discuten las dificultades para mantener la germinación de las semillas y almacenar semilla de forrajeras en el trópico húmedo.

Palabras clave: Almacenamiento de semilla, dormancia del embrión, dormancia física, dureza de semilla, trópico húmedo, viabilidad.

Introduction

Many tropical forage seeds produced and sold in Thailand are stored under ambient conditions in store rooms and shops where there is no control over temperature and humidity. The seeds are stored in conditions similar to those used to keep other grains for animal feed but which are not required to germinate. Forage seeds are sometimes carried over between years. There have been increasing concerns and reports about the declining germination quality of these forage seeds. In Australia, Hopkinson and English (2005) stored tropical grass seeds in a cool-room (10 °C and 50% relative humidity, RH) and found that germination rates of seeds initially with high viability remained high after 6 years cool-room storage. It was important for us to find the ideal storage conditions in Thailand that would maintain seed germination of our commercial forage seeds at acceptable levels for more than 1 year.

We undertook an experiment on the germination of commercial tropical forage seeds stored under ambient conditions or under conditions of controlled temperature and humidity. Species represented were Mulato II [Urochloa ruziziensis (syn. Brachiaria ruziziensis) x U. decumbens (syn. B. decumbens) x U. brizantha (syn. B. brizantha)], Mombasa guinea [Megathyrsus maximus (syn. Panicum maximum)], Tanzania guinea [M. maximus (syn. P. maximum)], Ubon paspalum (Paspalum atratum) and Ubon stylo (Stylosanthes guianensis). All are commercial lines that are produced and sold in Thailand.

The experiment commenced in January 2011. Germination results for the first 2 years (January 2011– January 2013) were reported in a previous paper (Hare et al. 2014). After 1 year of storage under ambient conditions, seeds of all grasses tested were almost dead. After 2 years cool-room storage (18–20 °C and 50% RH), germination percentage of Mombasa guinea, Tanzania guinea and Ubon paspalum seeds had not declined. We also found that apparent embryo dormancy and also physical dormancy in Mulato II and hardseededness in Ubon stylo persisted under storage. However, embryo dormancy in Mombasa and Tanzania guinea grasses was overcome within 6 months in cool-room storage (<u>Hare et al. 2014</u>).

We used a commercial seed store $(15 \times 7 \times 4 \text{ m})$ set at 18–20 °C and 50% RH. In this paper we report the performance of the initially tested seed lots under prolonged cool-room storage at temperatures which were higher than that used by <u>Hopkinson and English (2005)</u> but with similar humidity.

Materials and Methods

Seeds were harvested by village farmers from a number of villages in Northeast Thailand and Laos (Hare 2014) in October 2010 (Ubon paspalum 5,000 kg, Mombasa guinea 36,000 kg and Tanzania guinea 7,000 kg), November 2010 [Mulato II 12,000 kg: seed hand-knocked from seed heads (Hare et al. 2007a)] and January 2011 [Mulato II 16,000 kg and Ubon stylo 6,000 kg: seed swept from the ground (Hare et al. 2007a; 2007b)] and bulked within species, harvesting method and season. All harvested seeds were sun-dried to moisture levels in Table 1, cleaned and processed and entered storage in late January 2011. For the experiment, Mulato II seeds (handknocked and ground-swept) and the Ubon stylo seeds were divided into two 3 kg sublots before storage; the first sublot was scarified in sulphuric acid (96% normal) for 10 minutes, then washed and sun-dried to moisture levels in Table 1, while the second sublot was left untreated (unscarified). All seed lots and sublots consisted of 3 kg of seed drawn randomly from the total bulk of seed of each cultivar for the 2010/11 season, and placed into separate large (100 x 50 cm) commercial polyethylene bags, hand-tied tightly at the top.

The 3 kg bags of seeds consisting of one lot per bag were placed in 2 storage rooms, i.e. ambient conditions and a cool-room (Hare et al. 2014). The ambient seed

room was a storage shed at Ubon Ratchathani, Northeast Thailand (15° N, 104° E), where mean monthly temperatures were minimum 23 °C, maximum 34 °C and mean monthly RH was minimum 40%, maximum 92%. The cool-room was maintained at 18–20 °C and 50% RH throughout the study.

Seed samples were withdrawn from all storage lots in January of each year and tested for germination and moisture percentage. For each germination test, 3 replications of 100 seeds, randomly selected from each cultivar lot and sublot, were placed into covered petri dishes on filter paper wet with a 0.2% potassium nitrate solution and placed in a germination cabinet set to provide 16 h dark at 25 °C and 8 h light at 35 °C. The numbers of germinated seeds (normal seedlings), fresh ungerminated seeds or hard seeds, dead seeds and empty seeds were counted 7 and 14 days after wetting down. The ungerminated seeds were tested using the tetrazolium (TZ) assay test to determine if they were fresh ungerminated (dormant), hard or dead.

For germination testing of acid-scarified Mulato II seeds, further acid-scarification [sulphuric acid (96% normal) for 10 minutes] was conducted at testing on half the samples. To determine moisture percentage on each occasion, 3 samples of 10 g of seeds for each lot and sublot were weighed fresh and again after drying in an oven at 130 °C for 1 h (<u>ISTA 1993</u>). No seed moisture levels were measured in 2017.

Data from the experiment were subjected to analysis of variance using the IRRISTAT program from the International Rice Research Institute (IRRI). Each seed lot was analyzed separately with 7 years in storage as the treatments with 3 replications. The entry means were compared using Fisher's protected LSD ($P \le 0.05$).

Results

Moisture content

Moisture contents of seeds stored in the cool-room varied between 10.9 and 8.6% for the grasses and 8.3 and 5.1% for Ubon stylo (Table 1). Acid-scarified Mulato II seeds contained less moisture (9.1%) overall than untreated Mulato II seeds (9.8%). Mombasa guinea, Tanzania guinea and Ubon paspalum seeds averaged 9.9% seed moisture in cool-storage, similar to untreated Mulato II seeds (9.8%). Moisture level of untreated Ubon stylo seeds stored under ambient conditions was similar (5.5%) to that of untreated Ubon stylo seeds in cool-storage (5.2%).

Seed germination

Seeds of all grass cultivars maintained their germination for 2–3 years in cool-storage before germination started to decline steadily and dead and empty seeds increased (Table 2). After 6 years in cool storage, most seeds were either dead [Mulato II hand-knocked (Table 2), Mombasa and Tanzania guinea grasses (Table 3)], or had very low germination [Ubon paspalum 2% (Table 3)] or had less than 10% germination [Mulato II ground-swept 9% (Table 2)]. Only ground-swept Mulato II, that had been acid-scarified upon entering cool-storage and acid-scarified again when the germination test was conducted, gave a slightly better seed germination of 15% after 6 years in storage. The germination performance of Mulato II seeds, harvested by

Table 1. Effects of storage conditions on moisture contents of seeds of tropical forage cultivars during 2011–2016.

Cultivar	2011	2012	2013	2014	2015	2016
Cool-room ¹						
Mulato II ground-swept, acid-scarified ³	7.5	8.5	9.9	8.0	10.1	9.6
Mulato II ground-swept, unscarified4	10.6	8.8	10.2	8.6	9.4	9.8
Mulato II hand-knocked, acid-scarified	8.9	8.6	10.0	8.3	10.2	9.8
Mulato II hand-knocked, unscarified	10.5	9.3	10.7	9.0	10.8	10.2
Mombasa guinea	10.3	9.2	10.4	9.2	10.7	9.9
Tanzania guinea	10.1	9.0	10.4	8.9	10.4	9.7
Ubon paspalum	10.4	8.9	10.3	9.5	10.9	10.4
Ubon stylo acid-scarified	8.3	7.2	8.0	6.7	8.5	7.8
Ubon stylo unscarified	5.1	5.2	5.2	5.4	5.4	5.1
Ambient-room ²						
Ubon stylo acid-scarified	9.3	9.2 ⁵				
Ubon stylo unscarified	5.1	5.2	5.8	5.7	5.4	5.5

¹18–20 °C and 50% RH. ²Range in mean monthly temperatures - minimum 23 °C, maximum 34 °C; range in mean monthly RH - minimum 40%, maximum 92%. ³Scarified in sulphuric acid for 10 min, washed and dried. ⁴Not treated with acid. ⁵Seeds dead.

Table 2. Effects of cool-room storage conditions (18–20 °C and 50% RH) on germination of differently treated seeds of Mulato II hybrid brachiaria during 2011–2017.

Seed treatment	2011	2012	2013	2014	2015	2016	2017	LSD (P≤0.05)		
					germination					
Mulato II ground-swept, acid-scarified ¹	85	62	63	53	33	3	1	8.1		
Mulato II ground-swept, acid-scarified, more acid with test ²	90	90	89	84	75	42	15	8.4		
Mulato II ground-swept, unscarified ³	5	7	7	9	9	8	7	ns		
Mulato II ground-swept, unscarified, acid with test	84	75	81	79	65	40	9	8.1		
Mulato II hand-knocked, acid-scarified	70	63	68	20	19	1	0	17.9		
Mulato II hand-knocked, acid-scarified, more acid with test	86	82	84	62	46	8	0	13.0		
Mulato II hand-knocked, unscarified	0	1	1	3	4	1	1	ns		
Mulato II hand-knocked, unscarified, acid with test	51	75	86	61	41	3	0	10.3		
	Fresh ungerminated seeds (%)									
Mulato II ground-swept, acid-scarified ¹	11	29	27	31	42	39	14	3.6		
Mulato II ground swept, acid-scarified, more acid with test ²	8	9	8	1	5	4	1	2.2		
Mulato II ground-swept, unscarified ³	90	89	86	81	71	12	8	11.7		
Mulato II ground-swept, unscarified, acid with test	12	19	14	14	15	10	9	ns		
Mulato II hand-knocked, acid-scarified	28	25	18	12	10	5	0	3.2		
Mulato II hand-knocked, acid-scarified, more acid with test	10	11	10	8	4	4	0	4.5		
Mulato II hand-knocked, unscarified	97	91	89	40	8	0	0	3.3		
Mulato II hand-knocked, unscarified, acid with test	46	20	10	9	7	6	0	6.5		
				Dead and	empty see	ds (%)				
Mulato II ground-swept, acid-scarified ¹	4	9	10	16	25	58	85	7.3		
Mulato II ground swept, acid-scarified, more acid with test ²	2	1	3	15	20	52	84	9.1		
Mulato II ground-swept, unscarified ³	5	4	7	10	20	48	85	5.6		
Mulato II ground-swept, unscarified, acid with test	4	6	5	7	20	50	82	10.3		
Mulato II hand-knocked, acid-scarified	2	12	14	68	71	94	100	18.9		
Mulato II hand-knocked, acid-scarified, more acid with test	4	7	6	30	50	88	100	10.9		
Mulato II hand-knocked, unscarified	3	8	10	57	88	97	99	5.3		
Mulato II hand-knocked, unscarified, acid with test	3	5	4	30	52	91	100	10.3		

¹Scarified in sulphuric acid for 10 min, washed and dried. ²Scarified with sulphuric acid before storage and again before germination testing. ³Not treated with acid.

hand-knocking, deteriorated more quickly with time in storage than that of ground-swept Mulato II seeds (Table 2). After 4 years in storage, mean germination percentages of all lots of hand-knocked Mulato II seeds were below 50%, but it took 5 years in cool-storage for similar results to be reached with ground-swept Mulato II seeds.

Maximum seed germination of Mombasa guinea grass (68%) was reached after 1 year in cool-storage and those of Tanzania guinea grass (63%) and Ubon paspalum (85%)

after 2 years in cool-storage (Table 3). By the third year in cool-storage (2014), the germination of these 3 cultivars had declined rapidly to low levels (Table 3) and by the sixth year (2017), seeds were either dead (Mombasa and Tanzania) or had negligible germination (Ubon paspalum). The percentage of fresh ungerminated seeds for all cultivars quickly declined after the second year in cool-storage to levels well below 10% and the percentage of dead and empty seeds increased rapidly at the same time (Table 3).

Unscarified Ubon stylo seeds, when treated with acid at germination testing, maintained high germination percentages (>80%) for up to 5 years in both cool- and ambient-storage (Table 4). After 6 years in cool-storage, germination percentage of unscarified Ubon stylo seeds, treated with acid at the time of germination testing, was 3 times that of seeds acid-scarified following harvest (63 vs. 21%). Unscarified Ubon stylo seeds still displayed 45%

Table 3. Effects of cool-room (18–20 °C and 50% RH) storage conditions on germination of seeds of Mombasa guinea grass, Tanzania guinea grass and Ubon paspalum during 2011–2017.

Grass	2011	2012	2013	2014	2015	2016	2017	LSD (P≤0.05)	
	14-day germination (%)								
Mombasa guinea grass	35	68	65	27	14	7	0	9.8	
Tanzania guinea grass	43	56	63	30	31	12	0	11.1	
Ubon paspalum	73	79	85	51	37	7	2	7.2	
			F	resh unger	minated se	eds (%)			
Mombasa guinea grass	56	24	3	2	1	0	0	10.7	
Tanzania guinea grass	51	36	7	5	3	1	0	12.9	
Ubon paspalum	21	14	6	5	3	2	0	4.6	
				Dead and	empty seed	ds (%)			
Mombasa guinea grass	8	8	32	71	85	93	100	10.4	
Tanzania guinea grass	6	8	30	65	66	87	100	8.5	
Ubon paspalum	6	7	9	44	60	91	98	8.8	

 Table 4. Effects of storage conditions on germination of seeds of Ubon stylo during 2011–2017.

	2011	2012	2013	2014	2015	2016	2017	LSD (P≤0.05)
Cool-room ¹				14-day g	germination	n (%)		X
Ubon stylo acid-scarified ³	99	95	99	99	95	84	21	5.0
Ubon stylo unscarified ⁴	15	19	23	14	21	19	14	ns
Ubon stylo unscarified, acid with test ⁵	98	99	99	99	97	96	63	5.1
Ambient-room ²								
Ubon stylo acid-scarified	94	0^{6}						
Ubon stylo unscarified	10	3	2	1	2	2	3	5.2
Ubon stylo unscarified, acid with test	96	87	93	89	90	84	45	10.7
Cool-room ¹			I	Hard unger	minated se	eds (%)		
Ubon stylo acid-scarified ³	1	4	0	0	2	2	0	ns
Ubon stylo unscarified ⁴	85	81	76	84	76	71	7	8.6
Ubon stylo unscarified, acid with test ⁵	2	1	1	1	0	0	0	ns
Ambient-room ²								
Ubon stylo acid-scarified	6	0^{6}						
Ubon stylo unscarified	87	88	91	91	89	82	47	3.8
Ubon stylo unscarified, acid with test	3	5	2	5	4	2	3	ns
Cool-room ¹				Dea	d seeds (%)		
Ubon stylo acid-scarified ³	0	1	1	1	3	14	79	5.9
Ubon stylo unscarified ⁴	0	0	1	2	3	10	30	8.5
Ubon stylo unscarified, acid with test ⁵	0	0	0	0	3	4	37	5.3
Ambient-room ²					-			
Ubon stylo acid-scarified	0	0^{6}						
Ubon stylo unscarified	3	9	7	8	9	16	50	6.9
Ubon stylo unscarified, acid with test	1	8	5	6	6	14	52	5.6

¹18–20 °C and 50% RH. ²Range in mean monthly temperatures - minimum 23 °C, maximum 34 °C; range in mean monthly RH - minimum 40%, maximum 92%. ³Scarified in sulphuric acid for 10 min, washed and dried. ⁴Not treated with acid. ⁵Scarified with sulphuric acid before germination testing. ⁶Seeds all dead.

germination after 6 years in ambient-storage, when treated with acid at the time of germination testing. Ubon stylo seeds acid-scarified before entry into ambient storage, died after 1 year, but in cool-room storage maintained high germinations for 5 years (Table 4). Unscarified seeds in both cool- and ambient-storage, maintained high levels (>70%) of hardseededness for up to 5 years (Table 4).

Discussion

Germination percentages of forage grass seeds stored in a cool-room in this study varied substantially after 3 years of storage with many below 50% (which we arbitrarily define as minimal for sowing to ensure acceptable stands). The two guinea grass cultivars lost seed germination most rapidly to below 50% after 3 years coolroom storage, while Ubon paspalum seeds maintained higher germination for longer and could be kept in coolroom storage for up to 4 years before germination percentage dropped below 50%. The most durable grass seed was Mulato II with germination percentage remaining above 50% for longer than seed of the other grasses when stored in the cool-room: for 5 years if seed was harvested from the ground, but for only 4 years if seed was knocked out of the seed head at harvest. After 6 years in cool-storage seed of all grasses was either dead or had negligible levels of germination.

We define embryo dormancy as when seeds do not germinate but the embryo inside the seed is viable. We determined viability using the TZ assay test as it is the quickest test for evaluating seed viability. Embryo dormancy in seeds of Mombasa and Tanzania guinea grasses was overcome within 6 months in cool-room storage (Hare et al. 2014). On the other hand, acid scarification at the beginning of seed storage in 2011 was required to quickly overcome embryo dormancy of Mulato II seeds, but with time in cool-room storage, dormancy persisted and acid-scarified seeds had to be retreated with acid each year at the time of testing to get good germination. This secondary dormancy appears to be a physical type of dormancy, similar to that imposed by the tightly bound lemma and palea glumes over the caryopsis of unscarified seeds (Hare et al. 2008). In unscarified Mulato II seeds, aging in cool-storage did not overcome the physical dormancy attributable to these glumes, so seeds had to be acid-scarified at the time of the germination tests to achieve higher germination percentages (Table 2). Dormancy in unscarified Mulato II seeds is prolonged compared with that in other *Brachiaria* species, where dormancy has previously been measured to last only 10 months in *B. decumbens* seeds (Grof 1968) and up to 2 years in *B. dictyoneura* (now: *U. humidicola*) seeds (Hopkinson et al. 1996), while dormancy is inconsequential in *B. ruziziensis* (Hopkinson et al. 1996).

Seeds of Ubon stylo maintained high germination percentages (>85%) for up to 5 years when stored in ambient conditions, but only when they remained unscarified (Hare et al. 2014). This indicates that Ubon stylo seeds should not be scarified following harvest, if the aim is to store them for 1 year or more under ambient conditions. The situation differs in cool-storage, as Ubon stylo seeds, both acid-scarified and unscarified, maintained very high germination levels (>90%) for 4-5 years. Only in the sixth year did germination levels drop, particularly with acid-treated seeds, but they remained at levels above the low-to-zero germination levels of the grasses. Hardseededness, a type of physical dormancy, in Ubon stylo seeds was not overcome during either ambient- or cool-room storage and unscarified seeds in both storage rooms required treatment with acid each time a germination test was conducted to overcome hardseededness.

The moisture levels in grass seeds stored in the coolroom varied little from year to year, being above 10% in the first, third and fifth years of storage and 9% or less in the second, fourth and sixth years of storage. Since the bags of seeds in the study were moved around within the large cool-room, as commercial bags of seeds were introduced or withdrawn, possible variations in relative humidity in the room might have caused these moisture fluctuations. The seeds were stored in large commercial polyethylene bags and moisture exchange may have taken place. Seed life may have been extended if the seeds were dried to levels of 8% or less following harvesting and placed in sealed packages to prevent moisture exchange (Hopkinson and English 2005). However, the purpose of the study was to examine the life of our seed lots under commercial storage conditions (ambient- and coolstorage), so drying the seeds to very low seed moisture levels and packaging them in moisture-proof bags was considered impractical.

The results from this study and those from the first study (<u>Hare et al. 2014</u>) have important implications for the commercial storage and management of pasture seeds,

particularly grasses, in the humid tropics. Ideally, germination levels should be maintained above at least 60% for 12-15 months, until seed from the next season is ready for sale. Our first study showed that ambientstorage conditions in Thailand, even for a few months, were completely unsafe for seeds of our forage grasses, with a rapid decline in germination percentage to well below 50% within 8 months of entering storage (Hare et al. 2014). Typically, grass seeds in Thailand are harvested from October to January, cleaned, processed and placed in cool-room storage as soon as possible before the onset of the hot humid wet season in March. With this quick entry into cool-room storage, satisfactory germination levels (>70%) for most species are maintained prior to the key seed-purchasing period (March-October). The exception is Tanzania guinea seed, where it is difficult to obtain germination percentages of 70% under commercial conditions (60% is considered a satisfactory level).

Commercial seed lines rarely need to be stored for longer than 15 months in cool-storage. Conditions in the cool-room (18–20 °C and 50% RH) we used were satisfactory for seed storage prior to sale. <u>Hopkinson and English (2005)</u> found that the viability of grass seeds stored at 10 °C and 50% RH for 6–8 years remained constant. While the temperature in their room remained constant, we tested seeds in a commercial facility, where both temperature and humidity probably fluctuated (not measured) as the cool-room often remained open for 3 hours at a time to allow forklifts and trollies to enter and either deposit or remove seed.

It is important for traders who buy grass seeds (and then sell to third parties) to be made aware of the quick deterioration in viability of tropical grass seeds in ambient-storage. They should either sell the seeds within one month after purchase or, if seed is kept longer, keep it in air-conditioned rooms. Traders, for the most part, do not store grass seeds in cool conditions and many do not have access to air-conditioned storage rooms. Likewise, farmers should not buy grass seeds until they are almost ready to sow, unless they have an air-conditioned storage room. The rapid physiological deterioration of tropical grass seeds also has implications for shipping seeds internationally by sea in containers, as frequently grass seeds can be in transit by sea for 6-8 weeks; if hot and humid conditions exist, seeds should be shipped in refrigerated containers.

However, the quick deterioration in tropical grass seed germination has, to date, not limited the expansion of areas sown to improved pasture species in Southeast Asia and other humid tropical areas. Farmers may very well be increasing their seed sowing rates to allow for a possible decrease in germination. Some farmers may also conduct their own single germination tests before sowing to calculate seed sowing rates.

Acknowledgments

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Research Paper

Evaluation of growth parameters and forage yield of Sugar Graze and Jumbo Plus sorghum hybrids under three different spacings during the *maha* season in the dry zone of Sri Lanka

Efecto de distancia de siembra en el desarrollo y rendimiento de dos híbridos de sorgo forrajero (Sugar Graze y Jumbo Plus) durante la temporada de maha en la zona seca de Sri Lanka

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Abstract

A field experiment to evaluate the growth parameters and fodder yields of Sugar Graze and Jumbo Plus under occasional irrigation was conducted at 3 different plant spacings (30×15 , 30×45 and 30×60 cm) on a red-yellow latosol in the dry zone of Sri Lanka from August 2015 to January 2016. The design was a randomized block with 3 replications. Initial harvesting of fodder was done 60 days after planting and 2 ratoon yields were assessed at successive 60-day intervals. Plant spacing was inversely related (P<0.05) to dry matter (DM) yield with the narrowest spacing (30×15 cm) producing yields of 14.1 t DM/ha for Sugar Graze and 12.6 t DM/ha for Jumbo Plus at the initial harvest. Plant spacing also influenced leaf area, stem girth, root length and plant height in the initial harvest. Sugar Graze produced higher yields than Jumbo Plus at the initial and second ratoon harvests. Yields from ratoon crops were about 30% of those for the initial harvest. Further studies are needed to determine how these findings apply under the low-rainfall conditions of the *yala* season, and chemical analyses and animal feeding studies would provide valuable information on the nutritional value of the different forages.

Keywords: Dry matter yield, forage sorghum, ratoon crop, red yellow latosol, row width.

Resumen

En un latosol rojo-amarillo de la zona seca al norte de Sri Lanka entre agosto de 2015 y enero de 2016 se evaluaron algunas características de crecimiento y los rendimientos de forraje de los cultivares Sugar Graze y Jumbo Plus bajo riego ocasional usando 3 distancias de siembra $(30 \times 15, 30 \times 45 \text{ y } 30 \times 60 \text{ cm})$. Los tratamientos se dispusieron en un diseño de bloque al azar con 3 repeticiones. La primera cosecha de forraje se realizó 60 días después de la siembra, seguida por 2 cosechas de rebrote a un intervalo de 60 días cada una. Los resultados mostraron que la distancia de siembra se relacionó de manera inversa (P<0.05) con el rendimiento de materia seca (MS), siendo este más alto (14.1 t MS/ha) en la distancia 30×15 cm en la primera cosecha para el cv. Sugar Graze en comparación con el cv. Jumbo Plus (12.6 t MS/ha). La distancia de siembra también influyó en el área foliar, el grosor del tallo, la longitud de las raíces y la altura de la planta en la primera cosecha. Sugar Graze produjo mayores rendimientos que Jumbo Plus en la primera cosecha de rebrote. Los rendimientos en las dos cosechas de rebrote fueron de alrededor del 30% de la primera cosecha. Se requieren estudios adicionales para determinar cómo se comparan estos resultados con los que se puedan obtener en época seca (temporada *yala*). Además, análisis químicos y estudios nutricionales con animales proporcionarían información valiosa sobre el valor nutritivo de los diferentes forrajes.

Palabras clave: Distancia entre surcos, latosol amarillo-rojo, rebrote, rendimiento de materia seca.

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Introduction

High performance of farm animals, especially dairy cows, depends on the availability of adequate amounts of quality fodder and in developing countries, inadequacy of quality forage is the critical limitation to profitable animal production (Sarwar et al. 2002). Among the many options for overcoming the shortage of forage, the introduction of high-yielding crop varieties ranks highly (Bilal et al. 2001). However, in many developing countries, because of the ever-growing need for food for humans, only limited cultivated land can be allocated to produce fodder for livestock. Douglas (1980) recommended annual summer crops such as forage sorghum hybrids (*Sorghum* spp.) for use as alternative forage crops in drier areas in order to bridge the feed shortage gap.

Sugar Graze, a sweet sorghum × sweet sorghum hybrid, is a popular forage source among the livestock farmers of Sri Lanka and Jumbo Plus, a sweet sorghum \times Sudan grass hybrid, is still in the initial stages of introduction. Sugar Graze is a late-flowering cultivar with high yields, a crude protein (CP) concentration of 12-18% and a high sugar content that boosts feed quality and palatability, resulting in minimal feed wastage. In addition, the crop is resistant to a wide range of diseases. Mature Sugar Graze promotes good weight gains and provides adequate energy for livestock (Pacific Seeds 2009). Jumbo Plus, a forage sorghum hybrid cultivar, has excellent re-growth potential and high productivity and is adapted to both dryland and irrigated situations. It has similar CP concentration to Sugar Graze with 56-64% dry matter (DM) digestibility when the plant is 55-60 days old or at 5-10% flowering stage and can be used for grazing, silage making and rotational cropping (Forage Sorghum Guide 2015).

These crops have the potential to compete favorably with maize silage in terms of yield and nutritive value (Ketterings et al. 2005) and may be an appropriate alternative to maize for utilizing irrigation water in drought-prone areas. The shortage of ground water is the primary limitation to cultivating grass in the dry zone. As such, it is essential to select a drought-tolerant grass/fodder species, and Sri Lankan farmers cultivate fodder sorghum. In an initial study 7 cuttings were achieved from a single planting yielding 24 kg of fresh fodder/m² from a single cutting with plant spacing of 45×15 cm (Sivayoganathan 2016). While research on sorghum cultivars in Pakistan has shown marked differences between cultivars in green fodder yield and morphology under 30 cm row spacing (Bakhsh et al. 2015), similar data on forage sorghum hybrid

cultivars in the dry zone of Sri Lanka are limited. There is a need to assess the growth of these cultivars and how varying the plant spacing affects both yield and quality of forage so that the growing demand for forage by livestock can be met.

The present study was designed to determine the crop morphology, growth parameters and forage yield of Sugar Graze and Jumbo Plus under irrigation in the dry zone of Sri Lanka under 3 different plant spacings $(30 \times 15, 30 \times 45 \text{ and } 30 \times 60 \text{ cm})$.

Materials and Methods

This experiment was carried out at the livestock farm, Department of Animal Science, Faculty of Agriculture, University of Jaffna, Ariviyal Nagar, Kilinochchi (Figure 1), from August 2015 to January 2016. These months fall into Sri Lanka's *maha* season, i.e. the period September – February which experiences rainfall through the Northeast monsoon.

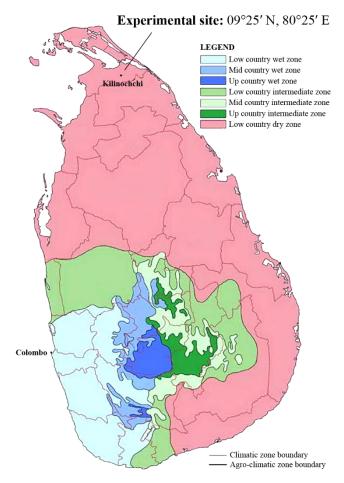


Figure 1. The experimental site, Kilinochchi District, in the dry zone of Sri Lanka.

The remaining period of the year is dry with the driest period being June to August. During the study period (August 2015–January 2016), rainfall in individual months varied greatly (Figure 2) and due to the extremely low water retention capacity irrigation had to be applied in some months.

According to Vavuniya meteorological data, the average monthly temperature in the region is 28.4 °C (range 25.6–30.0 °C), while maximum and minimum averages are 35.0 and 21.3 °C, respectively (Jaffna and Kilinochchi Water Supply and Sanitation Project 2010).

Soils of the area are red-yellow latosols (Haplustox), which are the most intensively cultivated soils of Jaffna Peninsula and have very low inherent fertility. Extremely poor water retention properties mean that dryland cropping is inappropriate, while conventional flood irrigation is impractical owing to very rapid infiltration and soil drying.

The experiment was laid out in a completely randomized design (CRD) in a factorial arrangement of 3 plant spacings $(30 \times 15, 30 \times 45 \text{ and } 30 \times 60 \text{ cm})$ and 2 cultivars (Sugar Graze and Jumbo Plus) with 3 replications. Sowing was on 5 August 2015.

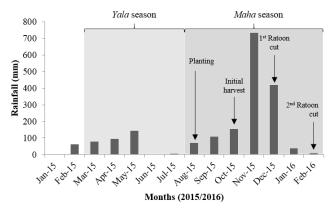


Figure 2. Monthly rainfall of Kilinochchi District from January 2015 to February 2016. Source: <u>Department of Census</u> and Statistics of Sri Lanka (2015); Kilinochchi District (2016).

Within rows spacing was kept constant at 30 cm and the spacing between rows was varied. Seeds were sown at the rate of 2 seeds per hill and seedlings were thinned to a single plant per hill 2 weeks after sowing resulting in plant populations of approximately 222,000, 74,000 and 55,000 plants/ha for inter-row spacings of 15, 45 and 60 cm, respectively. Cattle manure was applied at planting at the rate of 100 kg/ha (N: 1.2-1.9%, P: 0.2-0.5%, K: 0.5-1.1%) and inorganic fertilizers were applied 1 week after establishment of plants at the rate of 50 kg urea, 25 kg triple superphosphate and 12.5 kg muriate of potash/ha. During the dry spell of the study period, irrigation was done at weekly intervals. Plots were manually weeded at 30 days after planting to reduce competition from weeds.

Five plants were randomly selected from each plot for recording of leaf length and width, leaf area [leaf area factor (0.72 for forage sorghum) \times length \times width] (Arkel 1978), number of leaves per plant, stem girth, root length from the base of the plant to tip of the selected average lengthier rootlet, internodal elongation and plant height at weekly intervals. At 60 days after planting, on 3 October 2015, the crops were cut 15 cm from ground level and allowed to ratoon. Two ratoon cuts were made at 60-day intervals, on 1 December 2015 and 29 January 2016. Immediately after harvesting, fresh forage was weighed using a spring balance. Samples (approximately 2 kg) of the harvested forage from each experimental plot were selected and air-dried for 24 hours, followed by ovendrying at 75 °C for 72 h to constant weight for dry matter yield determination. Data were subjected to Analysis of Variance and mean separation was done with Duncan's multiple range test ($P \le 0.05$) (Duncan 1955) with SPSS (Statistical Package for Social Science) version 16.0 for Windows.

Results

Initial harvest

For the initial harvest the 2 cultivars generally responded differently to variation in plant spacing in terms of plant morphology (Table 1).

While leaf length, leaf width and leaf area were unaffected by plant spacing for Jumbo Plus, the narrow spacing $(30 \times 15 \text{ cm})$ produced narrower leaves with smaller area than the medium $(30 \times 45 \text{ cm})$ and wide $(30 \times 60 \text{ cm})$ spacings for Sugar Graze. Similarly, the narrow and wide spacings for Jumbo Plus produced more leaves per plant than the medium spacing, while the medium spacing for Sugar Graze produced more leaves than the narrow spacing. In general, the thickest stems were produced at the wide spacing and the thinnest at the narrow spacing.

A similar trend occurred with root length with longer root lengths generally being associated with wide plant spacing and shorter root lengths with narrow spacing. Plant height was unaffected by spacing for Jumbo Plus, while the wide spacing for Sugar Graze produced the tallest plants.

Overall, Sugar Graze displayed slightly longer, wider leaves with much greater area than Jumbo Plus but produced fewer leaves, though differences were not significant. This cultivar also produced longer roots than

Parameter		Sugar Graze		-	Jumbo Plus	
	30×15	30×45	30×60	30×15	30×45	30×60
Leaf length (cm)	$90.6 \pm 4.92a$	$91.4 \pm 2.17a$	$92.8 \pm 2.69a$	$86.5\pm3.52a$	$89.5 \pm 2.19a$	$87.6 \pm 3.11a$
Leaf width (cm)	$8.6\pm0.86b$	$9.2 \pm 0.80a$	$9.2 \pm 0.67a$	$7.27\pm0.64a$	$7.37\pm0.83a$	$7.15\pm0.91a$
Leaf area (cm ²)	$560\pm 64.5b$	$608 \pm 61.1a$	$616 \pm 48.8a$	$443 \pm 36.6a$	$463\pm 66.7a$	$473\pm67.9a$
Number of leaves/plant	$13.0\pm0.93b$	$13.9 \pm 0.83a$	$13.3 \pm 0.96 ab$	$15.0\pm0.93a$	$13.7\pm0.90b$	$15.1 \pm 1.68a$
Stem girth (cm)	$5.50\pm0.38b$	$6.23\pm0.86a$	$6.65\pm0.62a$	$5.65\pm0.52b$	$6.03 \pm 0.77 b$	$6.57\pm0.46a$
Root length (cm)	$23.5\pm0.01c$	$28.3\pm0.01a$	$26.0\pm0.01b$	$22.3\pm0.01c$	$24.3\pm0.01b$	$25.6\pm0.01a$
Internodal elongation (cm)	$21.9\pm2.54a$	$20.6 \pm 1.95a$	$21.4 \pm 2.41a$	$20.8\pm1.77a$	$22.7 \pm 1.16a$	$23.2 \pm 1.95a$
Plant height (cm)	$227\pm22.7b$	$239\pm40.6b$	$278 \pm 18.5a$	$290 \pm 14.8a$	$292 \pm 14.3a$	$297\pm25.4a$
Dry matter yield (t/ha)	$14.1\pm2.60a$	$11.4 \pm 1.94 b$	$11.3 \pm 1.80b$	$12.6\pm2.77a$	9.2 ± 2.81 ab	$6.3 \pm 1.71b$

Table 1. Effects of row spacing (30 x 15, 30 x 45, 30 x 60 cm) on growth parameters and yields of Sugar Graze and Jumbo Plus 60 days after planting (initial harvest).

Each value is a mean \pm SD for 3 replicates.

Within rows and cultivars, means without a common letter differ (P≤0.05).

Jumbo Plus but internodal elongation was greater for Jumbo Plus, resulting in taller plants.

Dry matter (DM) yields were inversely related to plant spacing with yield decreasing progressively as plant spacing increased, although differences were not always significant. Sugar Graze produced higher DM yields than Jumbo Plus (Table 1).

First ratoon crop

In the first ration crop some parameters, viz. leaf length, number of leaves and plant height, were not influenced by spacing in either cultivar, while leaf width, leaf area, internodal elongation and stem girth varied inconsistently with row spacing in the 2 cultivars (Table 2). Varietal differences also were noted among the morphological parameters of the first ration crop, where generally leaf length and width, leaf area, number of leaves per plant and stem girth were greater for Sugar Graze, while Jumbo Plus showed higher values for internodal elongation and plant height. Dry matter yields followed similar trends in both cultivars with declining yields as plant spacing increased, but differences were not always significant (Table 2). Varietal differences in DM yield were small.

Second ratoon crop

As for the first ratoon crop, row spacing had no significant effect on leaf width, leaf area and stem girth in either cultivar, while inconsistent responses occurred for the remaining morphological parameters (Table 3). There were consistent effects of row spacing on DM yields in both cultivars with yields declining as plant spacing increased, but differences were significant (P<0.05) only for Jumbo Plus. Dry matter yields for Jumbo Plus at the medium and wide spacings declined dramatically to about half those for Sugar Graze. Overall, increases in row spacing resulted in greater percentage yield decreases in Jumbo Plus than in Sugar Graze. Yields for both ratoon crops were generally about 25-35% of those obtained at the initial harvest.

Table 2. Effects of row spacing (30 x 15, 30 x 45, 30 x 60 cm) on growth parameters and yields of first ration crop of Sugar Graze and Jumbo Plus.

Parameter	-	Sugar Graze		-	Jumbo Plus	
	30×15	30×45	30×60	30×15	30×45	30×60
Leaf length (cm)	$78.3\pm5.07a$	$72.7\pm20.2a$	$78.1 \pm 3.93a$	$71.3 \pm 4.78a$	$71.4 \pm 5.28a$	$70.0 \pm 5.49a$
Leaf width (cm)	$6.79\pm0.70a$	$6.01\pm0.66b$	$6.37\pm0.52ab$	$4.19\pm0.41b$	$4.57\pm0.34a$	$4.45 \pm 0.49 ab$
Leaf area (cm ²)	$382\pm50.7a$	$317 \pm 100b$	358 ± 39.1ab	$215 \pm 29.4a$	$234 \pm 22.8a$	$224\pm35.6a$
Number of leaves/plant	$9.87\pm0.83a$	$9.53 \pm 1.25a$	$10.3\pm1.03a$	$9.33 \pm 1.18a$	9.53 ± 1.13a	$8.67 \pm 1.50 a$
Stem girth (cm)	$5.15\pm0.36a$	$4.45\pm0.34b$	$4.63\pm0.45b$	$3.83 \pm 0.37a$	$3.88 \pm 0.15a$	$3.63\pm0.21b$
Internodal elongation (cm)	$16.1 \pm 4.07a$	$17.8\pm2.62a$	$17.5 \pm 2.17a$	$16.9 \pm 3.32b$	$19.6 \pm 2.16a$	$21.1 \pm 1.18a$
Plant height (cm)	$115 \pm 19.9a$	$115 \pm 13.0a$	$119 \pm 14.6a$	$131 \pm 15.8a$	$138 \pm 11.6a$	$139 \pm 17.4a$
Dry matter yield (t/ha)	$3.36\pm0.531a$	$2.95\pm0.614a$	$2.36\pm0.416a$	$3.84\pm0.511a$	$3.27\pm0.309a$	$2.13\pm0.483b$

Each value is a mean \pm SD for 3 replicates.

Within rows and cultivars, means without a common letter differ ($P \le 0.05$).

Parameter		Sugar Graze	-		Jumbo Plus	
	30×15	30×45	30×60	30×15	30×45	30×60
Leaf length (cm)	$62.7 \pm 6.15a$	$59.0 \pm 3.58a$	$57.5 \pm 13.1a$	$60.7\pm5.57a$	$51.5 \pm 7.15b$	$54.5 \pm 4.63ab$
Leaf width (cm)	$5.78\pm0.78a$	$5.73\pm0.82a$	$5.65\pm0.98a$	$3.63 \pm 0.47a$	$3.68\pm0.62a$	$4.13\pm0.38a$
Leaf area (cm ²)	$262 \pm 50.7a$	$244\pm45.4a$	$239 \pm 89.0a$	$158 \pm 18.5a$	$137 \pm 35.0a$	$162 \pm 21.7a$
Number of leaves/plant	$7.00\pm0.63b$	$7.00\pm0.01b$	$8.33\pm0.52a$	$6.00\pm0.63a$	$6.00\pm0.63a$	$6.66\pm0.82a$
Stem girth (cm)	$4.50\pm0.55a$	$4.25\pm0.42a$	$4.08\pm0.38a$	$3.17 \pm 0.26a$	$3.20 \pm 0.32a$	$3.36 \pm 0.22a$
Internodal elongation (cm)	$20.0\pm2.83a$	$22.1\pm2.13a$	$19.0\pm1.95b$	$25.2\pm3.95a$	$26.7\pm3.36a$	$24.4\pm2.48a$
Plant height (cm)	$190 \pm 17.2a$	$178 \pm 10.2 ab$	$167 \pm 10.1b$	$191 \pm 9.12a$	$182\pm8.91a$	$168 \pm 9.31b$
Dry matter yield (t/ha)	$4.47 \pm 0.744a$	$3.29 \pm 1.090a$	$2.85\pm0.350a$	$4.33\pm0.358a$	$1.77\pm0.206b$	$1.42\pm0.539b$

Table 3. Effects of row spacing (30 x 15, 30 x 45, 30 x 60 cm) on growth parameters and yields of second ration crop of Sugar Graze and Jumbo Plus.

Each value is a mean \pm SD for 3 replicates.

Within rows and cultivars, means without a common letter differ ($P \le 0.05$).

Discussion

Leaf length and width

Leaf development has been described extensively for fodders, as growth is mostly reflected in large increases in leaf length as plants grow to maturity, accompanied by relatively small increases in width and thickness (Skinner and Nelson 1994). Large leaf lengths are also important for the survival of individual plants within a sward (Barre et al. 2015). Leaf length and width values observed for both cultivars during the present study were slightly greater than the values recorded by Singh et al. (2014) for leaf length (45–70 cm) and width (4–7 cm) of sorghum hybrids. Leaf length of Sugar Graze was similar to the 95 \pm 2.0 cm reported by Pahuja et al. (2014) in India, for the first cut at 50% flowering and a spacing of 15 × 45 cm, whereas leaf width was slightly higher than that recorded by the same authors (6 \pm 0.58 cm).

Leaf area

The results of the present study demonstrate that leaf area increases as plant spacing increases as shown by Lamana (2007). This could be due to less competition among plants for space and soil nutrients as the plant population per unit area decreased. Therefore, the lower population density which resulted from the wider plant spacing gives better conditions for more accumulation of photosynthetic products, better growth and expansion of foliage, which was in turn expressed in greater DM yields. The range of values for leaf area (440–615 cm²) for Sugar Graze and Jumbo Plus in the present study were in agreement with the values reported by Nabi et al. (2006) for advanced lines of forage sorghum cultivars. The higher mean leaf area in Sugar Graze (595 \pm 62.3 cm²) than in Jumbo Plus

 $(460 \pm 58.9 \text{ cm}^2)$ may be due to differences in genetic makeup of the cultivars. <u>Musa et al. (1993)</u>, <u>Naeem et al.</u> (2002), <u>Mahmud et al. (2003)</u> and Chohan et al. (2003; 2006) also observed variation in leaf area among various cultivars and varieties of forage sorghum.

Number of leaves per plant

The general absence of any consistent effect of row spacing on leaf number per plant is in agreement with the findings of <u>Liu et al. (2004)</u>, who observed for maize that it did not affect leaf number. In contrast <u>Lamana (2007)</u> reported that wider plant spacing in maize had a positive effect on number of leaves. The values recorded for number of leaves per plant for both cultivars in the present study were consistent with those of <u>Monteiro et al. (2012)</u>, who reported that number of leaves in forage sorghum is generally between 14 and 17. <u>Chohan et al. (2003)</u> and <u>Naeem et al. (2002)</u> also reported variation among different cultivars of sorghum for number of leaves per plant.

Stem girth

Stem girth recorded in the present study was similar to that reported by Pahuja et al. (2014) in India for stem girth of Sugar Graze (5.9 ± 0.21 cm at 50% flowering stage and 15×45 cm spacing). While <u>Yosef et al. (2009)</u> and <u>Ayub et al. (1999)</u> found significant variation in stem diameter among different cultivars of sorghum, cultivar differences in our study were not statistically significant (P>0.05).

Root length

The trend for root length to increase as row spacing increased would reflect greater competition between plants at the narrower spacings. Despite the shorter root length per plant at the narrow spacing, the much greater plant populations at this spacing would have resulted in substantially greater root length per unit area than at the wider spacings. As a result plants at the narrow spacing would have had better opportunity to utilize soil water and nutrients than at wider spacing, resulting in higher DM yields.

Internodal elongation and plant height

Plant height as a growth parameter is a result of elongation of the stem internodes, which is influenced by the environment as suggested by Weston (1967). In the current study taller plants were observed with wider spacing, which contrasts with reports in the literature that narrower spacing will give taller plants as a result of competition for sunlight (Lamana 2007). However, the absence of any effect of plant spacing on plant height of Jumbo Plus supports the finding of Roy and Biswas (1992) that plant height at maturity was not affected by plant spacing. The significant differences in plant height between the 2 cultivars may be due to genotypic variation, as differences in internodal elongation between varieties can lead to differences in height as reported by Evans (1975) and Weston (1967). Nabi et al. (2006) and Silungwe (2011), who worked with different forage sorghum cultivars, also reported plant heights (203-230 cm) lower than those in the present study (227–298 cm), as did Pahuja et al. (2014) for Sugar Graze $(189 \pm 1.9 \text{ cm})$ in India.

Dry matter yield

Plant spacing has a marked impact on the efficiency of use of land, light, water and nutrients. By optimizing plant spacing, highest yield potential can be achieved from the smallest possible area (Oseni and Fawusi 1986). The direct relationship between DM yield and plant population agrees with the findings of Fisher and Wilson (1975), who reported greater DM yield with higher plant populations than with lower plant populations. Wolf et al. (1993) and Graybill et al. (1991) also reported that DM yield of forage maize responded positively to plant density. This relationship would be affected by the availability of soil moisture, and the application of irrigation on a regular basis in this study would have ensured that all row spacings/plant populations had adequate water. Dry matter yield recorded for Sugar Graze in the current study seemed to be less affected by differences in row spacing than Jumbo Plus, which appeared not to be related to root length as there were no significant differences in root length between the

cultivars. Epasinghe et al. (2012) reported DM yields of Sugar Graze in Sri Lanka of 5,230 kg/ha at 60 days after planting at 45×15 cm spacing and these lower yields might be attributed to the differences in the spacing, soil fertility and environmental conditions. By contrast Nabi et al. (2006) recorded yields of 10,400–13,100 kg DM/ha for advanced lines of forage sorghums and Silungwe (2011) recorded 13,262 kg DM/ha at 15 cm row spacing 78 days after sowing for Sugar Graze.

Forage yield is a function of growth parameters, viz. plant population, plant height, leaf:stem ratio, leaf area, and leaf area index (Lamana 2007). The differences in DM yield between the 2 cultivars could be attributed to the fact that Sugar Graze exceeded Jumbo Plus in the growth parameters leaf length and width, leaf area and root length. Watson (1947) has shown that variation in total dry weight of plants is more dependent on variation in leaf area. Light interception capacity of the leaf is amplified with the increase in leaf area often leading to increase in photosynthesis and DM yield. Therefore, higher DM yield recorded for Sugar Graze might be attributed to its higher leaf area than Jumbo Plus.

First and second ratoon crops

The most consistent findings with the ration crops were that there were fewer leaves per plant, leaves were smaller, height was less and DM yields were lower than for the initial harvest. However, DM yield remained a factor of plant spacing with higher yields at narrower row spacing, indicating that plants were still accessing moisture and nutrients from the soil in sufficient quantities to maintain acceptable growth levels. The reduced yields are possibly a function of nutrient supply in the soil being depleted by the initial crop and a change in seasonal conditions over time. There were no significant differences between Sugar Graze and Jumbo Plus in DM yields for the first ratoon crop, in contrast with the generally higher yields for Sugar Graze in the initial crop and second ratoon crop. Despite having smaller leaves and thinner stems than Sugar Graze, the greater height of Jumbo Plus ensured that yields in the 2 cultivars were similar. The success of the second crop is often a function of how early the main crop was planted and harvested, which determines the seasonal conditions under which the first and second ratoon crops must grow. However, normally ratoon crops of sorghum are expected to yield from 25 to 35% of the main crop (Livingston and Coffman 1996), and our yields fall within this range. Significant differences in DM yield between main and ratoon crops have been reported by Saberi and Aishah (2014), when assessing yield responses of forage ration sorghum under varying salinity levels and irrigation frequencies.

Our findings suggest that both Sugar Graze and Jumbo Plus will grow satisfactorily under irrigation in this environment. While DM yields from the first harvest were excellent, yields from the ratoon crops were significantly lower despite the application of irrigation. A plant spacing of 30×15 cm produced the highest yields but results under rain-fed conditions would not necessarily be the same. Further studies to determine the performance in the low-rainfall (*yala*) season are necessary to determine desirable spacings under such dry conditions. Chemical analyses of forage and digestion studies would provide valuable information on the relative merits of these two cultivars for livestock feeding.

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Research Paper

Variation in carbohydrate and protein fractions, energy, digestibility and mineral concentrations in stover of sorghum cultivars

Variación en fracciones de carbohidratos y de proteína, energía, digestibilidad y concentraciones de minerales en rastrojos de cultivares de sorgo

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Abstract

The nutritional attributes of stover from 11 sorghum cultivars (SP 18005A x 220-2,3,6,7; PC-5; GGUB44 x SSG-59-3; ICSV-700; CSV-17; NRF-526; FM-1; SPV-1616; PVK-809; UPMC-503; and HC-308), selected on the basis of their diverse genetic backgrounds and use, were evaluated to aid in selecting parents superior in protein concentration and digestibility for use in sorghum breeding programs. Samples of stovers were collected after grain harvesting and analyzed. The CP concentrations in different cultivars differed (3.7-6.7%; P<0.05) as did NDF, ADF, cellulose and lignin concentrations (P<0.05). Total carbohydrate, non-structural carbohydrate and structural carbohydrate concentrations differed (P<0.05) amongst cultivars as did carbohydrate fractions (CA, CB1, CB2, CC; P<0.05). Protein fractions (P_{B1}, P_{B2}, P_{B3} and P_C) except P_A differed (P<0.05). Concentrations of stover protein fractions P_A and P_{B3} were lower than P_{B1} , P_{B2} and P_{C} . Unavailable protein fraction P_{C} was highest (P<0.05) in stover of SPV-1616 (36.8% CP) and lowest in ICSV-700 (20.4% CP). Concentrations of gross energy (GE), digestible energy (DE), metabolizable energy (ME) and total digestible nutrients (TDN) varied (P<0.05) and ICSV-700 had highest concentrations of DE, ME and TDN (2.60 kcal/g DM, 2.13 kcal/g DM and 59.0%, respectively). Energetic efficiency for maintenance (NE_M), lactation (NE_L) and growth (NE_G) differed (P<0.05) with ranges of 1.13–1.42, 0.41–0.70 and 0.95–1.33 kcal/g DM, respectively. Values for estimated DM intake, estimated digestible DM and relative feed value for stovers also varied (P<0.05) with ranges of 1.76–2.19%, 55.3–61.4% and 75.4–104.1%, respectively. In vitro dry matter digestibility was highest (P<0.05) for cultivars PVK-809 (55.7%) and ICSV-700 (54.3%). Macro- and micro-mineral concentrations also differed (P<0.05) across cultivar stovers. The wide genetic variability for nutritional attributes in stovers of sorghum cultivars indicates significant potential for improvement of stover quality through sorghum improvement programs, but care needs to be taken that grain and stover yields do not suffer.

Keywords: Energy values, nutritive value, sorghum stover, yields.

Resumen

En Hyderabad, India se evaluaron los atributos nutritivos de residuos de cosecha (rastrojo) de 11 cultivares de sorgo de grano (SP 18005A x 220-2,3,6,7; PC-5; GGUB44 x SSG-59-3; ICSV-700; CSV-17; NRF-526; FM-1; SPV -1616; PVK-809; UPMC-503; y HC-308), seleccionados por su diversidad genética y formas de uso, con el objeto de identificar líneas parentales superiores por concentración de proteína y digestibilidad, para uso eventual en programas de fitomejoramiento. Las concentraciones de proteína cruda difirieron entre los cultivares (3.7-6.7%; P<0.05) al igual que las concentraciones de NDF, ADF, celulosa y lignina (P<0.05). También difirieron (P<0.05) las concentraciones de carbohidratos totales, no estructurales y estructurales, y las fracciones de carbohidratos (C_A, C_{B1}, C_{B2}, C_C). Con excepción de P_A, las demás fracciones de proteína (P_{B1}, P_{B2}, P_{B3} y P_C) también difirieron (P<0.05). Las concentraciones de las

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fracciones proteicas P_A y P_{B3} fueron inferiores a P_{B1} , P_{B2} y P_C . La mayor (P<0.05) fracción de proteína no disponible (P_C) se encontró en el rastrojo de SPV-1616 (36.8% CP) y la más baja en ICSV-700 (20.4% CP). Las concentraciones de energía bruta, energía digestible, energía metabolizable y nutrientes digestibles totales (NDT) variaron entre los cultivares (P<0.05); ICSV-700 presentó las concentraciones más altas de energía digestible y metabolizable, y NDT (2.60 kcal/g MS, 2.13 kcal/g MS y 59.0%, respectivamente). La eficiencia energética para mantenimiento, lactancia y crecimiento difirieron entre los cultivares (P<0.05) con rangos de 1.13–1.42, 0.41–0.70 y 0.95–1.33 kcal/g de MS, respectivamente. El consumo estimado de MS, la MS digestible estimada y el valor relativo del alimento para los rastrojos también variaron (P<0.05) con rangos de 1.76–2.19%, 55.3–61.4% y 75.4–104.1%, respectivamente. La digestibilidad in vitro más alta de la MS (P<0.05) se encontró con los cultivares PVK-809 (55.7%) e ICSV-700 (54.3%). Las concentraciones de macro- y micro-minerales también variaron (P<0.05) entre cultivares. La amplia variabilidad genética de los atributos nutritivos en los rastrojos de los cultivares de sorgo indica un potencial significativo para mejorar la calidad del rastrojo a través de programas de fitomejoramiento, pero se debe considerar el riesgo de comprometer los rendimientos de grano y rastrojo.

Palabras clave: Calidad nutritiva, rendimientos, valor energético, variabilidad genética.

Introduction

Sorghum [Sorghum bicolor (L.) Moench] is one of the important cereal crops in the semi-arid tropics globally for providing human food, animal feed and raw materials for industrial use. In the present context of global climate change the crop is likely to become more important due to its adaptability to high temperature, water scarcity and saline conditions (Sanchez et al. 2002; Brouk and Bean 2011). Its tolerance of drought and saline conditions makes sorghum a valuable feed resource for growing on saline soils in arid and semi-arid regions (Fahmy et al. 2010).

India contributes 16% of global sorghum production and traditionally sorghum is grown both as fodder and grain crops in all states of India, with 3 southern states (Maharashtra, Karnataka and Andhra Pradesh) accounting for nearly 75% of sorghum's cultivable area and 85% of total sorghum production. It is grown as green fodder in the rainy season (July to mid-October, *Kharif* season) and later for grain as a food-feed crop.

Apart from producing grain as food for humans plus non-ruminant and ruminant livestock, sorghum residue (stover) is an important source of dry roughage for ruminants in the tropics, including India. The nutritive value of sorghum stover in terms of protein, energy and digestibility is low and stover is unable to provide a maintenance diet for ruminants. In view of the growing importance of crop residues for livestock feed, improving the nutritive value of sorghum stover is an important objecttive in the tropics (Rattunde et al. 2001). Blümmel and Reddy (2006) reported substantial variation in the fodder value of sorghum stovers and supported the concept of genetic enhancement to improve dual-purpose sorghum cultivars. Genetic variability in sorghum for various nutritional traits has been reported (Youngquist et al. 1990; Singh et al. 2014). There is a paucity of systematic information on nutritive value of improved forage sorghums for ranking of forage cultivars (<u>Akabari and Parmar 2014</u>) and also for selecting genetic material for use in sorghum improvement programs.

There is a need to quantify the genetic diversity of available sorghum cultivars in terms of nutritive value for use in breeding sorghum varieties or hybrids with higher stover value without compromising grain yield (<u>Rattunde 1998</u>; <u>Hash et al. 2000</u>). With this objective, a total of 11 sorghum cultivars were screened for variability in protein, carbohydrate and dry matter digestibility to select parents for subsequent use in sorghum breeding programs.

Materials and Methods

Production, sampling and processing of sorghum stovers

Eleven sorghum cultivars (SP 18005A x 220-2,3,6,7; PC-5; GGUB44 x SSG-59-3; ICSV-700; CSV-17; NRF-526; FM-1; SPV-1616; PVK-809; UPMC-503; and HC-308), selected on the basis of diverse genetic backgrounds, use and yield (stover and grain; Table 1) were grown at the research farm of Indian Institute of Millet Research, Hyderabad, India, in a randomized block design with 3 replications in plots of 5 x 4 m spaced at 45 cm between rows and 15 cm between plants within rows. A basal dose of 80 kg N and 40 kg P/ha was applied, with a further 40 kg N/ha 30 days after sowing. The variation in number of days to grain ripening since planting varied among cultivars: CSV-17 matured in 100 days and ICSV-700 matured in 122 days with the remainder intermediate. Yields of grain and stover were measured following grain harvesting and a composite stover sample was taken from each replication of individual cultivars for chemical analysis. The stover samples were dried in a hot-air oven at 60–65 °C for 96 h to constant weight. Dried samples were then ground through a 1-mm sieve using an electrically operated Willey mill and subsequently stored in plastic containers for laboratory analysis.

Chemical analyses

Dry matter (DM), crude protein (CP), ether extract (EE) and ash concentrations of sorghum stover samples were estimated as per procedures of <u>AOAC (2000)</u>. Fiber fractions, namely neutral detergent fiber (NDF), acid detergent fiber (ADF), cellulose and lignin, were determined following the detergent method of <u>Van Soest et</u> <u>al. (1991)</u> using Fiber Tech analyzer (FibraPlus FES 6, Pelican, Chennai, India). Heat-labile α -amylase and sodium sulphite were not used in NDF solution. Lignin (sa) was determined by dissolving cellulose with sulfuric acid in the ADF residue (<u>Van Soest et al. 1991</u>). Cellulose was estimated as the difference between ADF and lignin (sa) in the sequential analysis and hemicellulose was calculated as difference between NDF and ADF concentrations.

Carbohydrate and protein fractions

Total carbohydrates (tCHO) of stover samples were calculated as 100 - (CP + EE + ash). Carbohydrate fractions in the samples were estimated as per Cornell Net Carbohydrate and Protein (CNCP) system (Sniffen et al. 1992), which classifies carbohydrate fractions according to degradation rate into 4 fractions, viz. CA - rapidly degradable sugars; CB1 - intermediately degradable starch and pectin; C_{B2} - slowly degradable cell wall; and C_C unavailable/lignin-bound cell wall. Structural carbohydrates (SC) were calculated as the difference between NDF and neutral detergent insoluble protein (NDIP), while non-structural carbohydrates (NSC) were estimated as the difference between tCHO and SC (Caballero et al. 2001). Starch in samples was estimated by extracting stover samples in 80% ethyl alcohol to solubilize free sugars, lipids, pigments and waxes. The residue rich in starch was solubilized with perchloric acid and the extract was treated with anthrone-sulfuric acid to determine glucose colorimetrically using glucose standard (Sastry et al. 1991). A factor of 0.9 was used to convert glucose into starch (mg %).

The CP of stover samples was partitioned into 5 fractions according to the Cornell Net Carbohydrate and Protein System (CNCPS; <u>Sniffen et al. 1992</u>) as modified by <u>Licitra et al. (1996</u>). Neutral detergent insoluble protein (NDIP), acid detergent insoluble protein (ADIP) and non-protein nitrogen (NPN) were estimated following

the standard method (Licitra et al. 1996). For NDIP and ADIP, samples extracted with neutral detergent and acid detergent solutions, respectively, were analyzed as Kjeldahl N x 6.25 using semi-auto analyzer (Kel Plus Classic-DX Pelican India). For NPN estimation, samples were treated with sodium tungstate (0.30 molar) and filtered, and residual nitrogen was determined by the Kjeldahl procedure. Non-protein nitrogen of the sample was calculated by subtracting residual nitrogen from total nitrogen. Soluble protein (SP) was estimated by treating the samples in borate-phosphate buffer, pH 6.7-6.8, consisting of monosodium phosphate (Na₂PO₄.H₂O) 12.2 g/L, sodium tetraborate (Na₂B₄O₇.10H₂O) 8.91 g/L and tertiary butyl alcohol 100 mL/L and freshly prepared 10% sodium azide solution (Krishnamoorthy et al. 1983). The N estimated in the residue gives the insoluble protein fraction. The SP was calculated by subtracting insoluble protein from total CP.

Intake, digestibility, energy, feed value

To calculate DM intake (DMI), digestible dry matter (DDM), relative feed value (RFV), total digestible nutrients (TDN) and net energy (NE) of the stovers for different animal functions, i.e. lactation (NE_L), weight gain (NE_G) and maintenance (NE_M), equations given by <u>Undersander et al.</u> (1993) were used. Digestible energy (DE) and net energy (NE) values were calculated using equations of <u>Fonnesbeck et al.</u> (1984) and <u>Khalil et al.</u> (1986), respectively. The in vitro dry matter digestibility (IVDMD) was estimated using the 2-stage technique of <u>Tilley and Terry</u> (1963) by incubating 0.5 g of sample in inoculum of sheep maintained on a mixed grass hay-concentrate diet.

Minerals

Samples of sorghum stovers were wet-digested with 3:1 HNO₃:perchloric acid mixture, cooled and filtered through Whatman 42 filter paper. The aliquot was used for estimation of calcium (Ca), copper (Cu), zinc (Zn), iron (Fe), cobalt (Co) and manganese (Mn) using an atomic absorption spectrophotometer (Varian AA 240) against their standards. Phosphorus was estimated colorimetrically using Bartor's reagent according to <u>AOAC</u> (2000).

Statistical analysis

Data were subjected to analysis of variance of SPSS 17.0 to test the differences between sorghum cultivars for chemical composition, carbohydrate and protein fractions, energy values, digestibility and mineral concentrations.

Variable means were compared for significance at P<0.05 level (<u>Snedecor and Cochran 1994</u>).

Results

Grain and stover yields

Stover yields in the various cultivars varied from 7.61 t/ha (CSV-17) to 13.7 t/ha (SP 18005A x 220-2,3,6,7), while grain yields ranged from 1.59 t/ha (FM-1) to 4.51 t/ha (SPV-1616) (Table 1).

Chemical composition

All chemical parameters varied (P<0.05) between cultivars. Crude protein was highest in SP 18005A x 220-2, 3, 6, 7 and PC5 (6.6 and 6.7%, respectively) and lowest in UPMC-503 (3.7%; Table 2). The OM and EE concentrations in stovers varied (P<0.05), with ranges of 91.0–93.5% and 1.05–1.61%, respectively. NDF ranged from 55.0% (ICSV-700) to 68.2% (CSV-17), ADF from 35.3% (ICSV-700) to 43.1% (CSV-17), cellulose from 27.9% (ICSV-700) to 33.8% (CSV-17) and lignin from 4.33% (PVK-809) to 5.79% (CSV-17) (P<0.05).

Carbohydrate fractions

Concentrations of tCHO, NSC and SC of sorghum stovers differed (P<0.05) between cultivars (Table 3). Total carbohydrates varied from 88.6% (UPMC-503) to 83.3% (SP 18005A x 220-2,3,6,7), while structural carbohydrates were highest in CSV-17 (66.4%) and lowest in ICSV-700 (53.6% DM). Similarly the carbohydrate fractions (C_A, C_{B1}, C_{B2}, C_C) differed significantly (P<0.05) across the sorghum cultivars. The highly degradable carbohydrate fraction (C_A) was highest (P<0.05) in stover of ICSV-700 (30.3%) and lowest in CSV-17 (16.7%). On the other hand the slowly degradable carbohydrate fraction (C_{B2}) was lowest in ICSV-700 (53.8%) and highest in CSV-17 (66.4%).

Protein fractions

The protein fractions P_{B1} , P_{B2} , P_{B3} and P_C differed significantly (P<0.05) in stovers of the sorghum cultivars (Table 4). Lignin-bound/unavailable protein fraction P_C was highest (P<0.05) in stover of SPV-1616 (36.8%) and lowest in ICSV-700 (20.4% CP).

Table 1. Sorghum cultivars used in the study, their use and yields of stover and grain.

Cultivar	Commodity/Major utility	Stover yield (t/ha)	Grain yield (t/ha)
SP 18005A x 220-2,3,6,7	Sweet sorghum/ High biomass	13.7a	2.82cd
PC-5	Fodder	8.96bc	2.23de
GGUB44 x SSG-59-3	Fodder	10.05abc	2.18d
ICSV-700	Sweet sorghum/ High biomass	12.51ab	2.7c
CSV-17	Grain & fodder	7.61c	3.4c
NRF-526	Sweet sorghum/ High biomass	12.07ab	2.46d
FM-1	Fodder	9.49abc	1.59e
SPV-1616	Grain & fodder	11.34abc	4.51a
PVK-809	Grain & fodder	10.76abc	3.89ab
UMPC-503	Fodder	8.6c	2.03de
HC-308	Fodder	9.95abc	1.79e

Means followed by different letters within columns differ significantly at P<0.05 level.

 Table 2. Chemical composition (% DM) of stover from 11 sorghum cultivars.

Variable	SP 18005A	PC-5	GGUB44 x	ICSV-700	CSV-17	NRF-526	FM-1	SPV-1616	5 PVK-809	UPMC-503	HC-308	sem	Sig
	x 220-2,3,6,7		SSG-59-3										-
СР	6.6ef	6.71f	5.87de	4.88bc	4.53abc	4.43abc	5.03c	3.87ab	4.46abc	3.68a	4.07ab	0.134	< 0.0001
OM	91.5abc	93.1de	93.0de	93.2de	92.6cde	91.9abcd	93.3de	91.1a	91.0a	93.5e	92.4bcde	0.159	< 0.0001
EE	1.21ab	1.14ab	1.24ab	1.05a	1.28abc	1.61d	1.51d	1.25ab	1.29cd	1.14ab	1.22ab	0.026	< 0.0001
NDF	63.0b	64.0b	62.3b	55.0a	68.2c	62.1b	61.5b	61.7b	62.0b	63.9b	64.1b	0.474	< 0.0001
ADF	38.1ab	38.7b	36.8ab	35.3a	43.1c	38.9b	36.2ab	37.0ab	38.0ab	37.7ab	39.0b	0.335	< 0.0001
Cellulose	30.3b	31.7b	29.9ab	27.9a	33.8c	30.7b	29.4ab	30.2b	30.8b	31.5b	31.1b	0.251	< 0.0001
Hemicellulose	25.5bc	25.6bc	25.5bc	19.7a	25.2bc	23.3b	25.4bc	24.7bc	23.9bc	26.2c	25.1bc	0.286	< 0.0001
Lignin	5.51ef	4.84abc	4.48ab	4.96bcde	5.79f	5.58ef	4.73abc	4.54ab	4.33a	4.64abc	5.04bcd	0.074	< 0.0001

Means followed by different letters within rows differ significantly at P<0.05 level.

CP - crude protein; OM - organic matter; EE - ether extract; NDF - neutral detergent fiber; ADF - acid detergent fiber.

Variable	SP 18005A x	PC-5	GGUB44 x	ICSV-700	CSV-17	NRF-526	FM-1	SPV-1616	PVK-809	UPMC-503	HC-308	sem	Sig
	220-2,3,6,7		SSG-59-3										
tCHO (% DM)	83.3a	84.9ab	85.1ab	86.8cd	86.3bc	85.3ab	86.6cd	85.8abc	84.6abc	88.6d	87.0cd	0.306	0.007
NSC (% DM)	22.7a	23.1a	24.4a	33.2b	19.9a	24.8a	27.6ab	26.1ab	25.7ab	26.5ab	24.8a	0.760	0.102
SC (% DM)	60.7b	61.8bc	60.7b	53.6a	66.4c	60.5b	59.0b	59.7b	58.9b	62.1bc	62.2bc	0.631	0.012
CA (% tCHO)	20.2ab	20.2ab	22.7ab	30.3c	16.7a	21.6ab	24.5bc	20.9ab	21.9ab	21.7ab	20. 9ab	0.744	0.002
C _{B1} (% tCHO)	0.95a	2.26bc	1.60abc	1.50abc	1.41ab	1.38ab	2.20bc	4.30d	3.57d	3.64d	2.55c	0.188	0.028
C _{B2} (% tCHO)	62.8b	64.5b	64.0b	53.8a	66.4b	61.1b	59.9b	61.4b	62.2b	61.9b	62.5b	0.680	0.0001
Cc (% tCHO)	16.0d	13.0ab	11.7a	14.4bcd	15.5cd	15.9d	13.3abc	13.3ab	12.3ab	12.7ab	14.2bcd	03.05	0.063
C _{B1} (% tCHO) C _{B2} (% tCHO)	62.8b 16.0d	64.5b	64.0b	53.8a	66.4b 15.5cd	61.1b	59.9b 13.3abc	61.4b	62.2b	61.9b	62.5b	0.680	0.0

 Table 3. Carbohydrate and its fractions in stovers of 11 sorghum cultivars.

Means followed by different letters within rows differ significantly at P<0.05 level.

tCHO - total carbohydrates; NSC – non-structural carbohydrates; SC - structural carbohydrates; C_A - rapidly degradable sugars; C_{B1} - intermediately degradable starch and pectins; C_{B2} - slowly degradable cell wall; C_C - unavailable/lignin-bound cell wall.

Energy and its efficiency for animal functions

Energy value in terms of GE, DE, ME and TDN in stovers differed significantly (P<0.05; Table 5). Cultivar ICSV-700 had highest concentrations of DE, ME and TDN (2.60 kcal/g DM, 2.13 kcal/g DM and 59.0%, respectively), while CSV-17 had the lowest (2.16 g/kg DM, 1.77 kcal/g DM and 48.9%, respectively). The energetic efficiency for different animal functions, viz. NE_M, NE_G and NE_L, also differed (P<0.05) amongst the sorghum cultivars, with ranges of 1.13–1.42, 0.41–0.70 and 0.95–1.33 kcal/g DM, respectively.

Intake, digestibility and relative feed value

The calculated values of DMI, DDM and RFV for stovers of the 11 sorghum cultivars varied significantly (P<0.05;

Table 6) with ranges of 1.76-2.19%, 55.3-61.4% and 75.4-104.1%, respectively. In vitro dry matter digestibility (IVDMD) of stovers was highest (P<0.05) for cultivars PVK-809 (55.7%) and ICSV-700 (54.3%) and lowest for CSV-17 (40.3%).

Macro- and micro-minerals

Macro- and micro-mineral concentrations in stovers differed (P<0.05) across sorghum cultivars (Table 7). Stover from SPV-1616 had lowest Ca and P concentrations (216 and 39.9 mg/kg, respectively) with highest Ca in NRF-526 (398 mg/kg) and highest P in HC-308 (71 mg/kg). The concentrations of micro-minerals, viz. Cu, Zn, Fe, Mn and Co, ranged between 1.47 and 9.59, 14.2 and 35.5, 109 and 281, 46.5 and 112.5, and 1.74 and 5.44 ug/g, respectively.

Table 4. Protein fractions (% CP) of stovers from 11 sorghum cultivars.

Variable	SP 18005A x	PC-5	GGUB44 x	ICSV-700	CSV-17	NRF-526	FM-1	SPV-	PVK-	UPMC-503	HC-308	sem	Sig
	220-2,3,6,7		SSG-59-3					1616	809				-
PA	8.95	9.28	6.66	7.73	8.55	6.44	8.49	6.94	11.51	10.29	9.15	0.48	0.661
\mathbf{P}_{B1}	26.7ab	26.1ab	21.8a	25.1ab	26.2ab	26.6ab	25.3ab	25.4ab	22.9a	30.0bc	34.1c	0.66	0.010
\mathbf{P}_{B2}	33.1bc	30.2abc	36.6c	28.8abc	28.5abc	33.7c	21.4ab	25.0abc	20.9a	21.5ab	20.8a	1.23	0.040
P_{B3}	4.99a	12.93abc	11.30abc	17.96c	12.30abc	12.17abc	16.58bc	5.79a	11.03ab	9.67ab	7.82a	0.854	0.016
Pc	26.3ab	21.5a	23.6a	20.4a	24.4a	21.1a	28.3ab	36.8c	33.6bc	28.6ab	28.5ab	0.999	0.002

Means followed by different letters within rows differ significantly at P<0.05 level.

 P_A - non-protein nitrogen; P_{B1} - buffer-soluble protein; P_{B2} - neutral detergent-soluble protein; P_{B3} - acid detergent-soluble protein; P_C - indigestible protein.

Variable	SP 18005A x	PC-5	GGUB44 x	ICSV-700) CSV-17	NRF-526	FM-1	SPV-1616	PVK-809	UPMC-503	3 HC-308	sem	Sig
	220-2,3,6,7		SSG-59-3										
GE (kcal/g)	4.17bc	4.01a	4.11abc	4.04ab	4.12abc	4.14abc	4.22c	4.16abc	4.04ab	4.13abc	4.13abc	0.014	0.118
DE (kcal/g)	2.44bc	2.41b	2.52bc	2.60c	2.16a	2.40b	2.55bc	2.50bc	2.44bc	2.46bc	2.39b	0.019	<.0001
ME (kcal/g)	2.00bc	1.90b	2.07bc	2.13c	1.77a	1.97b	2.10bc	2.06bc	2.01bc	2.02bc	1.96b	0.016	<.0001
TDN (%)	55.3bc	54.6b	57.1bc	59.0c	48.9a	54.4b	57.9bc	56.8bc	55.4bc	55.9bc	54.2b	0.437	<.0001
NE _L (kcal/g)	1.19bc	1.16b	1.26bc	1.33c	0.95a	1.15b	1.29bc	1.24bc	1.19bc	1.21bc	1.15b	0.016	<.0001
NE _G (kcal/g)	0.59bc	0.57b	0.65bc	0.70c	0.41a	0.57b	0.67bc	0.64bc	0.60bc	0.61bc	0.56b	0.013	<.0001
NE _M (kcal/g)	1.31bc	1.29b	1.37bc	1.42c	1.13a	1.29b	1.39bc	1.36bc	1.32bc	1.33bc	1.28b	0.0126	<.0001

Table 5. Energy and energetic efficiency for different animal functions of 11 sorghum stovers.

Means followed by different letters within rows differ significantly at P<0.05 level.

GE - gross energy; DE - digestible energy; ME - metabolizable energy; TDN - total digestible nutrients; NE_L - net energy for lactation; NE_G - net energy for growth/gain; NE_M - net energy for maintenance.

Table 6. Predicted dry matter intake, digestibility and feed value of stovers from 11 different sorghum cultivars.

18005A x	PC-5	GGUB44 x I	CSV-700	CSV-17	NRF-526	FM-1	SPV-1616	PVK-809	UPMC-503	HC-308	sem	sig
20-2,3,6,7		SSG-59-3										-
51.1cde	47.6bc	52.6def	54.3ef	40.3a	47.7bc	53.7ef	50.9cde	55.7f	48.4bcd	45.7b	0.552	<.0001
59.2bc	58.7b	60.2bc	61.4bc	55.3a	58.6b	60.7bc	60.1bc	59.3bc	59.5bc	58.5b	0.261	<.0001
1.89b	1.86ab	1.93b	2.19c	1.76a	1.94b	1.95b	1.95b	1.94b	1.88b	1.87ab	0.015	<.0001
86.7b	85.1b	90.1b	104.1c	75.4a	88.2b	92.0b	90.9b	89.7b	87.0b	85.0b	1.038	<.0001
2	20-2,3,6,7 51.1cde 59.2bc 1.89b	20-2,3,6,7 51.1cde 47.6bc 59.2bc 58.7b 1.89b 1.86ab	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 59.2bc 58.7b 60.2bc 1.89b 1.86ab 1.93b	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 59.2bc 58.7b 60.2bc 61.4bc 1.89b 1.86ab 1.93b 2.19c	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 59.2bc 58.7b 60.2bc 61.4bc 55.3a 1.89b 1.86ab 1.93b 2.19c 1.76a	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 53.7ef 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 60.7bc 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b 1.95b	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 53.7ef 50.9cde 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 60.7bc 60.1bc 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b 1.95b 1.95b	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 53.7ef 50.9cde 55.7f 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 60.7bc 60.1bc 59.3bc 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b 1.95b 1.95b 1.94b	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 53.7ef 50.9cde 55.7f 48.4bcd 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 60.7bc 60.1bc 59.3bc 59.5bc 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b 1.95b 1.94b 1.88b	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 53.7ef 50.9cde 55.7f 48.4bcd 45.7b 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 60.7bc 60.1bc 59.3bc 59.5bc 58.5b 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b 1.95b 1.94b 1.88b 1.87ab	20-2,3,6,7 SSG-59-3 51.1cde 47.6bc 52.6def 54.3ef 40.3a 47.7bc 53.7ef 50.9cde 55.7f 48.4bcd 45.7b 0.552 59.2bc 58.7b 60.2bc 61.4bc 55.3a 58.6b 60.7bc 60.1bc 59.3bc 59.5bc 58.5b 0.261 1.89b 1.86ab 1.93b 2.19c 1.76a 1.94b 1.95b 1.95b 1.94b 1.88b 1.87ab 0.015

Means followed by different letters within rows differ significantly at P<0.05 level. IVDMD - in vitro dry matter digestibility; DDM - estimated digestible dry matter; DMI - estimated dry matter intake; RFV - relative feed value.

 Table 7. Macro- and micro-mineral concentrations in stovers of 11 sorghum cultivars.

Variable	SP 18005A x	PC-5	GGUB44 x	ICSV-700	CSV-17	NRF-526	FM-1	SPV-1616	PVK-809	UPMC-503	HC-308	sem	Sig
	220-2,3,6,7		SSG-59-3										
Ca (mg/kg)	343c	236bc	259ab	241ab	341ab	398cd	228bc	216a	215a	241ab	285abc	10.01	0.001
P (mg/kg)	45.9abc	42.3ab	62.6abc	56.4abc	47.9abc	47.2abc	60.7abc	39.9ab	42ab	65.6bc	71c	2.60	0.071
Mg (mg/kg)	58.6	49.8	44.5	46.0	42.9	54.5	52.1	44.9	45.0	42.9	48.6	2.40	0.013
Cu (ug/g)	4.45b	1.86a	1.55a	1.54a	5.45b	8.51c	8.25c	1.47a	2.94a	3.71a	9.59c	1.76	0.032
Zn (ug/g)	14.9	17.2	16.4	27.3	32.2	18.2	14.2	24.5	28.6	35.5	23.8	0.623	0.410
Fe (ug/g)	230ab	277b	281b	195ab	241ab	272b	173ab	149a	164ab	109a	126a	20.17	0.001
Mn (ug/g)	98.3cd	69.2abc	112.5d	68.3abc	54.6ab	71.3abc	54.7ab	74.3abc	83.4bcd	65.0abc	46.5a	3.94	0.011
Co (ug/g)	3.86abc	3.06abc	4.30bc	3.50abc	3.04abc	4.85bc	1.74a	3.05abc	2.57ab	4.25bc	5.44c	0.258	0.026

Means followed by different letters within rows differ significantly at P<0.05 level.

Discussion

Grain and stover yields

The stover yields of high biomass lines SP 18005A x 220-2,3,6,7, ICSV-700 and NRF-526 were higher, but not significantly so, than those of fodder and grain types SPV-1616 and PVK-809. This is expected because the high biomass lines were specially bred for higher biomass. On the other hand, the grain yields were higher in SPV-1616 and PVK-809 followed by CSV-17. The former two varieties were bred for maximizing grain yield with superior stover yield. <u>Umakanth et al. (2012)</u> observed that SPV 1616 showed high adaptability for grain and

fodder yields and biomass, and hence better suited as a dual purpose sorghum variety. <u>Sharma (2013)</u> observed that CSV 17 was a good grain yielding variety that had least stover yield in western Rajasthan, India.

Chemical composition

Cereal stovers and straws are usually low in crude protein and rich in fiber concentrations, unable even to meet the minimum CP requirements (7%) for maintenance of animals and rumen microbes (Minson 1990), so there is need to supplement these stovers with protein rich leguminous forage or non-protein nitrogen or protein sources. In the present study CP concentrations (3.7– 6.7%) of sorghum stovers are below the maintenance requirement for ruminants. Mativavarira et al. (2013) reported that CP concentrations of stovers varied (P<0.05) across cultivars and ranged between 5.6 and 6.6%, which supports our findings. Varietal differences for sorghum stover quality have been reported for protein and cell wall concentrations (Badve et al. 1993). Fiber fractions, viz. NDF, ADF, cellulose and lignin, are in general agreement with the earlier recorded values of Elseed et al. (2007) across 5 sorghum varieties. Crude protein, OM and EE concentrations of sorghum stovers reported by Misra et al. (2009) were on par with our results, while their NDF and ADF concentrations were higher than our values. Like the present study, variability in NDF, ADF, cellulose and lignin concentrations of sorghum stovers in different cultivars has been reported earlier (Garg et al. 2012; Hamed et al. 2015).

Carbohydrate and protein fractions

Carbohydrates constitute the main energy source of plants (50-80%) and play an important role in animal nutrition as a prime source of energy for rumen microorganisms (Van Soest 1994). In our study total carbohydrate concentrations of sorghum stovers varied between 83.4 and 88.6% DM, and exceeded the 78.5% DM reported by Das et al. (2015). Carbohydrate accumulation in fodder crops is influenced by several factors like plant species, variety, growth stage and environmental conditions during growth (Buxton and Fales 1994). Concentrations of SC and NSC differed (P<0.05) across the cultivars as suggested by Ferraris and Charles-Edwards (1986) and McBee and Miller (1990). Swarna et al. (2015), while evaluating the nutritive value of crop residues, found that C_A, C_{B1}, C_{B2} and Cc concentrations in sorghum stover were 14.7, 1.12, 56.8 and 28.0% of tCHO levels, a pattern of carbohydrate fractions identical with our results. Relatively low C_C values (11.7-16.0% tCHO) in our study may be due to the lower lignin concentrations in our stovers than in theirs. In our results carbohydrate fraction C_{B2} was highest in CSV-17 (66.4%) and lowest in ICSV-700 (53.8% tCHO). This is probably a function of the higher NDF and hemicellulose concentrations in CSV-17 and lower NDF and hemicellulose concentrations in ICSV-700. This was substantiated by the fact that forage with high NDF levels had higher concentrations of the CB2 fraction, which is more slowly degraded in the rumen, impacting microbial synthesis and animal performance (Ribeiro et al. 2001). Higher hemicellulose concentrations result in higher concentrations of carbohydrate C_{B2} fraction. <u>Carvalho et al. (2007)</u> reported that NDF concentration influences carbohydrate fraction C_{B2} and forages high in NDF concentration usually have higher values of C_{B2} . Values of carbohydrate fraction C_C in our study (11.7–16.0 % tCHO) were generally lower than the 15.8–25.2% reported by <u>Malafaia et al. (1998)</u> for grasses.

Protein fractions (P_{B1} , P_{B2} , P_{B3} and P_C) differed (P<0.05) across sorghum cultivars, which may be attributed to differences in concentrations of CP and lignin. About 5–15% of total forage N is bound to lignin, or rather, is unavailable to ruminal microorganisms (Van Soest 1994). Protein fraction P_C of stovers recorded in our study ranged between 20.4 and 36.8% CP, exceeding the above levels, probably due to variability in lignin concentrations. Forages, fermented grains and byproduct feeds contain significant amounts of fraction P_{B3} (Krishnamoorthy et al. 1983).

Energy and its efficiency

Energy density of roughages is a primary parameter influencing animal productivity. Stovers from the evaluated sorghum cultivars had adequate energy, except for CSV17 (ME 1.77 kcal/g), to meet the maintenance requirement of livestock (ME 2.0 kcal/g DM recommended for ruminants; ICAR 2013). The DE and ME concentrations in our study differed (P<0.05) across cultivars, being highest for ICSV-700 (2.60 and 2.13 kcal/g DM) and lowest for CSV-17 (2.16 and 1.77 kcal/g DM). The range of values for DE (2.16-2.6 kcal/g DM)and ME (1.77-2.13 kcal/g DM) are similar to the 2.14-2.51 kcal DE/g DM and 1.76-2.05 kcal ME/g DM recorded by Neumann et al. (2002), the 1.70-2.00 kcal ME/g DM reported by Garg et al. (2012) and the 1.6–1.72 kcal ME/g DM reported by Mativavarira et al. (2013). The variation in TDN concentrations in our study (59.0% for ICSV-700 to 48.9% for CSV-17) is a function of differences in fiber concentrations, as fiber is often used as a negative index of nutritive value in the prediction of total digestible nutrients and net energy. Sorghum stover TDN concentrations of 46.5-56.5% reported by Garg et al. (2012) cover a similar range to our findings, while Beef Magazine (2015) suggests TDN concentrations of sorghum stover are about 54% and Neumann et al. (2002) reported TDN of silage made from sorghum hybrids between 54.4 and 62.2%. Studies on the net energy efficiency of sorghum stovers for animal production functions is limited and values for NE_M, NE_G and NE_L reported in Beef Magazine (2015) for sorghum stover of 1.06, 0.40 and 1.06 kcal/g DM corroborate our results.

Mean values of NE_M, NE_G and NE_L reported by <u>Bean et al. (2011)</u> for hay made from the second cut of 32 sorghum hybrids were 1.13, 0.59 and 1.21 kcal/g DM, i.e. within the range of energy values for sorghum stovers recorded in our study.

Intake, digestibility and relative feed value

From a livestock production view point, intake and digestibility are the main criteria in breeding programs for quality improvement in most cereal fodder crops. Dietary fiber concentration, its digestibility and rate of degradation in the rumen are the most important forage characteristics that determine DMI (Roche et al. 2008). The differences in predicted DMI levels we recorded (1.76-2.19%) may be attributed to differences in NDF concentrations. The NDF concentration of CSV-17 was 68.2%, which exceeds the 60.0% usually considered as the threshold likely to significantly reduce intake in ruminants (Zewdu 2005). Mahanta and Pachauri (2005) recorded DMI between 1.84 and 2.55% for sheep fed silage from 3 sorghum cultivars ad lib. Relative feed value of hay from second cut of 32 sorghum hybrids ranged between 106 and 126 (Bean et al. 2011), which exceeded the 75.4-100 we recorded. We attribute the lower RFV of stovers in the present study to their lower quality relative to the whole plants examined at a younger age by Bean et al. (2011), i.e. higher NDF and ADF concentrations as these influence the intake and digestibility of a fodder. Forage containing 41% ADF and 53% NDF is considered to have an RFV of 100 and RFV values decrease as the concentrations of NDF and ADF increase with crop maturity.

The variability in digestibility values may be attributed to differences in cell wall concentrations. Elseed et al. (2007) reported effective degradability of dry matter of stovers from different cultivars between 44.4 and 67.7%, which covers a similar range to our IVDMD and DDM values. Bani et al. (2007) recorded an inverse relationship between forage fiber fractions and DM digestibility, while Barriere et al. (2003) and Seven and Cerci (2006) indicated that nitrogen concentration and cell wall polysaccharides determine the digestibility of a crop. The IVDMD of sorghum stover of 53.3% reported by Misra et al (2009) is consistent with our stover IVDMD values. The lower concentrations of NDF, cellulose and lignin in ICSV-700 and FM-1 could explain their higher IVDMD and DDM values (Tovar-Gomez et al. 1997; Zerbini and Thomas 2003), while the highest lignin concentration (5.79%) in stover of sorghum cultivar CSV-17 may explain the lowest IVDMD and DDM values for this cultivar.

Macro- and micro-minerals

Forages neither contain all the required minerals nor are they present in adequate quantity to meet animal requirements (Vargas and McDowell 1997). Calcium and phosphorus constitute the major portion (up to 70%) of the body's total mineral elements, play a vital role in almost all tissues in the body and must be available to livestock in proper quantities and ratio (McDowell et al. 1993). The Ca concentrations that we found, 215–343 mg/kg, should fulfill the maintenance requirements of ruminants (270-570 mg/kg; NRC 2001), but P and Mg concentrations in stovers were low (39.9-71 and 42.9-58.6 mg/kg) and unable to meet the critical levels (220 and 120–220 mg/kg) recommended for ruminants. While the Ca concentrations in sorghum stover/straws reported by <u>Ramesh et al. (2014)</u> and <u>Garg et al. (2003)</u> are more or less similar to our values, P concentrations reported by these workers are higher than our values. Misra et al. (2015) reported P and Mg concentrations in sorghum stovers (N = 31) similar to ours. The concentrations of Cu (1.47-9.59 ug/g), Zn (14.2-35.5 ug/g) and Fe (109-281 ug/g) recorded in our study were within sorghum stover values reported by Ramesh et al. (2014) and Misra et al. (2015). The low concentrations of many minerals in straws and stovers are probably due to maturity and possible transfer of nutrients to seeds. Mineral concentrations in feeds and fodders are influenced by a number of factors (soil pH, soil type, plant species, stage of growth and harvest, crop yield, intensity of agriculture system, climate, fertilizer rate etc. (British Geological Survey 1992; McDowell et al. 1993).

The results from this study revealed significant variability in apparent nutritive value of the sorghum stovers tested. This indicates that there is considerable potential for selecting appropriate genotypes to include in breeding programs to improve stover quality. While stovers of all genotypes had adequate energy to meet ruminant maintenance requirements, protein concentrations were low and quite variable. While there is potential to improve stover quality by breeding, care would need to be taken to ensure grain and stover yields did not suffer as a result. Feeding studies with animals would throw more light on the predicted feed intakes and digestible dry matter values reported in this study.

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Comunicación breve

Evaluación de un sistema de manejo de *Axonopus catarinensis* en rotación basado en el remanente de forraje no pastado (Renopa)

Long-term assessment of a new rotational-grazing management strategy called PUP-grazing (proportion of un-grazed pasture)

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Resumen

Una nueva estrategia de pastoreo rotativo – Renopa (remanente no pastado) – fue comparada productiva y económicamente con el pastoreo rotativo tradicional (PRT) en pasturas de *Axonopus catarinensis* durante dos periodos (2013/14 y 2015/16) en la provincia de Misiones, Argentina, utilizando terneros de cruza Cebú. El promedio del remanente no pastado del Renopa y el PRT fue 11.5 y 3.4% del área de pastura, respectivamente. La ganancia diaria de peso fue significativamente más alta (P<0.05) para el Renopa que para el PRT (606 vs. 420 g/día). La ganancia de peso por hectárea también fue 35% más alta para el Renopa (194 vs. 144 kg/ha por periodo). El ingreso bruto por hectárea fue mucho más alto para el Renopa (US\$ 85.7 vs. 8.4/ha por periodo). Concluimos que el Renopa tiene un alto potencial para mejorar la productividad de pasturas de *A. catarinensis*.

Palabras clave: Consumo de forraje, ganado vacuno, ganancia de peso, manejo del pastoreo, margen bruto.

Abstract

A new rotational-grazing management strategy called PUP-grazing (proportion of un-grazed pasture, which is the estimated percentage of pasture vegetation without signs of being consumed) was compared with the traditional rotational-grazing management strategy (TGMS, which is based on residual sward height) using Brahman cross steers on *Axonopus catarinensis* over two periods (2013/14 and 2015/16) in Misiones, Argentina. The proportion of un-grazed pasture for PUP and TGMS was 11.5 and 3.4%, respectively, of the pasture area. Average daily liveweight gain/animal was significantly higher for PUP than for TGMS (606 vs. 420 g/d; P<0.05) while liveweight gain per hectare was 35% greater for PUP (194 vs. 144 kg/ha/period). The gross margin per hectare was much higher for PUP than for TGMS (US\$ 85.7 vs. 8.4/ha/period). These results indicate that on *A. catarinensis* pastures PUP-grazing has the potential for greater animal and economic performance than the TGMS.

Keywords: Cattle, forage consumption, grazing management, gross margin, liveweight gain.

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Introducción

Una práctica de manejo común en sistemas de pastoreo rotativo consiste en asignar la permanencia de los animales en potreros en función de la altura del forraje residual. No obstante esta estrategia es limitada por el hecho que la altura residual a la cual el consumo de forraje y la respuesta animal decrecen depende no solo de la especie de pastura sino también de la altura al comienzo del pastoreo (<u>Benvenutti et al. 2016; 2017</u>).

Benvenutti et al. (2016; 2017) en pasturas del pasto Jesuita Gigante (Axonopus catarinensis) en un sistema de rotación y en caña de azúcar, encontraron que aplicando el criterio del forraje remanente no-pastado (Renopa) mejoró el consumo de forraje por bovinos en pasturas con diferentes alturas. En estos trabajos se encontró que al comienzo del periodo de ocupación los animales alcanzaron el mayor nivel de consumo diario de forraje especialmente en el estrato superior de hojas de la pastura. El consumo se redujo cuando más del 90% de la superficie de la pastura fue defoliada por primera vez, debido a que los animales no tuvieron otra opción que consumir el estrato inferior de esta de menor calidad con alta proporción de tallos y material senescente. Con base en esta observación se determinó que el mejor indicador para el cambio de potrero de los animales sin pérdida de consumo de forraje era la proporción del remanente no-pastado, la cual fue equivalente entre el 5 y 10% del total del área del potrero, independiente de la especie de pastura y la altura al comienzo del pastoreo (Benvenutti et al. 2016; 2017). Este remanente estuvo normalmente asociado con sectores de la pastura contaminados por heces.

Tomando como base las observaciones anteriores, durante dos periodos, 2013/14 y 2015/16, en Misiones, Argentina, en un experimento de más largo plazo, utilizando variables productivas y económicas, se evalúo el sistema Renopa con el objeto de determinar si el mayor consumo de forraje observado resulta en una mayor respuesta productiva y económica, cuando se compara con el pastoreo rotativo tradicional (PRT).

Materiales y Métodos

Periodo 2013/14

Quince días antes del comienzo del ensayo, animales jóvenes Cebú cruzados alimentados previamente con caña de azúcar fueron sometidos a un periodo de acostumbramiento en una pastura de *Axonopus catarinensis*.

El área experimental consistió en 2 ha bajo árboles de *Pinus taeda*, la cual fue dividida en dos partes iguales y

cada una de ellas en ocho subpotreros de 1,250 m² cada uno. En una de las partes se tomaron las observaciones con el sistema Renopa y en la otra con el PRT. Los animales entraron a cada tratamiento el 6 de noviembre de 2013, con pesos promedio de 194 ± 2.6 kg y 190 ± 3.2 kg para el Renopa y el PRT, respectivamente. Las evaluaciones se hicieron durante un periodo de 105 días.

En el sistema PRT los animales fueron cambiados de subpotrero cuando por efecto del consumo animal, la pastura alcanzó una altura promedio en 28 mediciones diarias de 20 cm sobre el nivel del suelo.

En el caso del Renopa los animales fueron cambiados de potrero cuando el porcentaje del remanente de pastura fue aproximadamente el 10% del total del área del subpotrero. Para ello cada día se hicieron mediciones en dos transectos diagonales y se contaron el número de pasos realizados sobre pastura no pastada (Foto 1). Cuando este número estuvo entre 5 y el 10% del total de pasos se procedió al cambio del subpotrero. Por ejemplo, si el total de pasos en ambas diagonales fue de 200, y se registraron entre 10 y 20 pasos en sitios no pastados, los animales fueron cambiados de potrero.

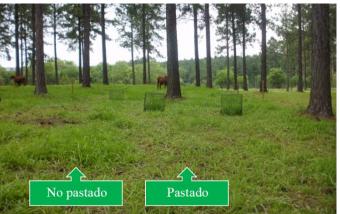


Foto 1. Área experimental de *Axonopus catarinensis* en un sistema silvopastoril con *Pinus taeda*, con sectores pastados y no pastados.

Utilizando un septómetro Decagon se determinó el porcentaje relativo de luz incidente bajo árboles. Para ello se tomaron 400 mediciones en cada tratamiento bajo árboles y 200 en sitios sin cobertura de árboles (100 antes y 100 después de las mediciones bajo arboles). Las mediciones se hicieron entre las 12:40 y las 12:52 horas, cuando el sol se encontraba en el zenit. Los porcentajes de radiación a cielo abierto (sin árboles) fueron 49% para Renopa y 48.5% para el PRT. Este nivel de luz cercano a 50% en ambos sistemas se considera suficiente para un proceso fotosintético aceptable de la pastura, por lo que se decidió continuar el experimento en estas condiciones.

Periodo 2015/16

Entre el 9 de septiembre de 2015 y el 18 de febrero de 2016 se realizó un segundo ensayo de 162 días de evaluación para comparar el PRT y el Renopa en un lote diferente de *A. catarinensis* de 2.4 ha bajo un rodal de *P. taeda* de 40 años de edad. Cada mitad del lote, 1.2 ha, fue dividida en cinco subpotreros con una superficie promedio de 2,400 m² cada uno para evaluar ambos sistemas en forma independiente. En este caso se repitió el protocolo empleado en el ensayo del periodo 2013/14. Las mediciones mostraron que la luz incidente era muy baja (28%) por lo que se realizó un raleo de los árboles hasta un nivel aproximado de 50% de luz incidente. A diferencia del periodo 2013/14, en este segundo periodo se midieron la producción y utilización del forraje siguiendo el método descrito por <u>Benvenutti et al. (2016)</u>.

Análisis estadístico

Los resultados fueron analizados por varianza utilizando el programa Genstat 2016. Este análisis fue realizado con medidas repetidas en el tiempo utilizando las múltiples mediciones de ganancia de peso vivo animal realizadas en cada periodo. En el contexto de este experimento que fue repetido en dos periodos, la interacción entre periodo y tratamiento es considerado como efecto aleatorio (Welham et al. 2014). Por ello los periodos de evaluación se consideraron como repeticiones de los tratamientos de pastoreo.

Resultados y Discusión

Ganancia de peso vivo

En el periodo experimental los animales en el sistema Renopa ganaron 35% más de peso vivo (P<0.05) que en el PRT (194 vs. 144 kg/ha) (Cuadro 1).

En el Cuadro 2 se observa la probable causa de la ganancia superior de peso vivo en el sistema Renopa, en comparación con PRT. En el Renopa, la proporción de forraje remanente no pastado fue aproximadamente 11%, lo que permite que el consumo voluntario y por tanto la ganancia diaria de peso no sean limitados antes del cambio de potrero, como se observó en los estudios anteriores (Benvenutti et al. 2016; 2017). El mayor consumo voluntario de los animales en el sistema Renopa es debido al mejor acceso al estrato superior de la gramínea (= hojas con valor nutritivo más alto) por los animales. En contraste, en el sistema PRT los animales acceden al estrato inferior (= hojas y tallos de menor valor nutritivo) resultando un remanente de 3%, lo cual produce una caída marcada en el consumo voluntario de forraje de menor calidad previo al cambio de subpotrero.

Cuadro 1. Peso vivo (PV) por animal y ganancia de peso por animal/día (GDP) en los sistemas Renopa y PRT en dos periodos de evaluación.

Periodo	Sistema de pastoreo	PV inicial (kg)	PV final (kg)	Ganancia de peso (kg/anim.)	GDP (g/día)	Ganancia de peso (kg/ha)
2013/14 (105 días)	Renopa	194	260	66	629	198
	PRT	190	238	47	451	142
2015/16 (162 días)	Renopa	184	279	95	584	189
	PRT	187	250	63	389	145
Promedio	Renopa	189	270	80	606	194
	PRŤ	189	244	55	420	144
Significancia		>0.05	< 0.05	< 0.05	< 0.05	< 0.05

Cuadro 2. Altura de pasturas después del pastoreo y remanente no pastado.

Periodo	Sistema de pastoreo	Altura después del pastoreo (cm)	Remanente no pastado (% area)
2013/14	Renopa	32.5	11.6
	PRT	21.2	2.9
2015/16	Renopa	32.7	11.3
	PRŤ	19.5	3.9
Promedio	Renopa	32.6	11.5
	PRT	20.4	3.4
Significancia		< 0.05	< 0.05

Cuadro 3. Promedios (kg/ha) de pastura disponible y residual, y pastura utilizada para los tratamientos Renopa y PRT en el periodo	
2015/16.	

Sistema	Pastura disponible antes del pastoreo	Pastura residual	Pastura utilizada ¹	Pastura utilizada total ²
Renopa	1,650	1,008	642	8,346
PRT	1,502	893	608	7,299

¹Calculada como la diferencia entre la pastura disponible menos la residual.

²Calculada como la sumatoria de la pastura utilizada para todos los cambios de potreros durante el periodo.

Cuadro 4. Determinación y comparación de los márgenes brutos¹ para Renopa y PRT.

Concepto	Renopa				PRT			
	Periodo 2013/14 Perio		Periodo 2015/16		Periodo 2013/14		Periodo 2015/16	
	US\$/ha	US\$/anim.	US\$/ha	US\$/anim.	US\$/ha	US\$/anim.	US\$/ha	US\$/anim.
Ingresos brutos	1,275.51	425.17	912.37	456.18	1,167.60	389.20	943.45	408.83
Gastos por compras	1,129.02	376.34	713.88	356.94	1,105.74	368.58	837.14	362.76
Suplementación mineral	3.30	1.10	3.40	1.70	3.30	1.10	3.92	1.70
Sanidad	18.20	6.07	12.13	6,.07	18.20	6.07	14.00	6.07
Mano de obra	77.46	25.82	59.02	29.51	77.46	25.82	68.10	29.51
Margen bruto	47.53	15.84	123.94	61.97	-37.10	-12.37	20.30	8.79

¹El precio de compra del ganado fue estimado en 1.94 US\$/kg vivo y de venta en 1.72 US\$.

Disponibilidad y utilización de forraje

Los datos en el Cuadro 3 muestran que en el sistema Renopa la celeridad de rebrote de la pastura fue mayor y por tanto también la producción y utilización de MS (8.34 t/ha) que en el PRT (7.29 t/ha). Esta mayor producción es probablemente debido al mayor área foliar remanente en el sistema Renopa.

Cabe destacar que para lograr el nivel deseado de remanente no pastado hay que considerar tanto la carga animal como el tamaño de potrero. En un potrero pequeño, un grupo grande de animales tal vez ni deja una planta no pastada el primer día de pastoreo, mientras unos pocos animales en un potrero grande tal vez nunca alcanzan a comer el estrato superior en el 90–95% del área de la pastura (para dejar un 5–10% deseable de remanente no pastado).

Además, en pasturas con gramíneas de hábito erecto o cespitoso de porte alto como *Panicum maximum*, el Renopa puede dejar un nivel alto de residuos (<u>Benvenutti et al. 2017</u>) con la tendencia a aumentar con cada pastoreo. Para evitar la acumulación indeseada de residuos se pueden usar varias estrategias tales como evitar que la pastura este muy alta al momento del pastoreo, o utilizar un segundo rodeo de animales y/o una máquina para consumir o cortar el residuo después del pastoreo del rodeo principal.

Evaluación económica

El sistema Renopa generó mayor disponibilidad de forraje, mayor tiempo de pastoreo, mayor carga de peso vivo, mayor GDP y por consiguiente mejor terminación y mayor producción de carne por hectárea, lo cual resultó en mayor margen bruto que en el PRT (Cuadro 4). Se puede apreciar que el sistema Renopa produjo una mayor eficiencia económica respecto del PRT, en ambos periodos.

Conclusión

Las observaciones y resultados en este trabajo sugieren que el sistema Renopa, que es una estrategia simple y útil para definir, con base en la proporción de forraje no pastado o remanente en la pastura, cuándo mover los animales en un sistema de pastoreo rotativo en *A. catarinensis*, tiene un alto potencial para maximizar la producción animal, el crecimiento de la gramínea y el retorno económico del sistema.

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