DANIEL DA CUNHA

AGRONOMIC TRAITS AND ADAPTATION OF THE CROPGRO-PERENNIAL FORAGE MODEL TO PREDICT THE GROWTH OF FIVE *Brachiaria* GENOTYPES

LAVRAS – MG
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Thesis presented to the Federal University of Lavras, as part of the Animal Science Graduate Program requirements, area of Ruminant Production and Nutrition, to obtain the Ph.D. title in Animal Science.

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Approved on February 25, 2022.

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To my dad Carlos Roberto and grandfather Norberto Martins
(in memorian) who inspire me and are a reference for good attitudes,
character, dedication, and love for the family.

To my mother Rosângela, who spares no effort to see my
happiness and personal and professional evolution.

To my wife Ana Luiza for all the support, love, and trust.

I dedicate

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"It is not coincidence, and it is not luck, I know it comes from above." I really want to thank God, because in his presence I learned to see miracles in miracles. Even with the losses, God held me tight and gave me victories, healings, and countless miracles. He also opened doors that no one could close. And it was only by His grace that I managed to get here. He gave me the opportunity to live with my father and grandfather long enough to learn to be a man of principle and learn to take care of the family. He also gave me the opportunity to learn from my mother how to be a good son and from my grandmother how to be a good grandchild. When I left Mom's house, He gave me the opportunity to learn from many others, such as Advisors, Teachers, Collaborators and Friends. And I made the best of each of those opportunities. He gave me the chance to meet Dr. Márcio, my advisor who has helped me a lot so far. And after so many attempts, God also gave me the chance to study abroad and meet amazing people like Dr. Boote and Dr. Hoogenboom who have kept me motivated even in difficult times, and until now. He also gave me the opportunity to make amazing friends like Fabio, Maurício and Camila, Vinícius and Vanessa. All friends I hope to take from Florida for life. I cannot fail to mention my friend Sérgio, who also helped me and always surprises me by making me see things in a different way. In the meantime, God has also given me the opportunity to meet and build my own family. Again, God working miracles upon miracles, where we can not see the possibilities. With my wife Ana Luiza, who deserves the greatest credit for giving me strength and helping me to overcome the challenges I faced during this period and that I will face from now on. And, to give me the opportunity to learn and live with my mother and brother-in-law. Each one contributing their respective part to bring me here, with patience, affection, dedication and love. Finally, I thank God for putting me in front of excellent Professors, Researchers and Employees of the Department of Animal Science, Plant Physiology and Forest Sciences, as they were part of my training and indirectly contributed to this work. To the many friends I have made before and during this journey, because you have helped me to overcome obstacles with my head held high. To CAPES (Coordination for the Improvement of Higher Education Personnel) for granting the national and international scholarship and for the Postgraduate Program in Animal Science at UFLA.

To God and to all of you, my thanks.

"Everyone wants to live on top of the mountain, but all the happiness and growth occurs while you are climbing it."

(Andy Rooney)

"All things are difficult before they are easy."

(John Norley)

ABSTRACT

Brachiaria spp. play a key role in animal production for grassland-based systems in tropical and subtropical regions. The evaluation of forage productivity and canopy characteristics allows us to understand pasture responses over time and identify high-yield genotypes with different defoliation management needs. Our objectives were to compare and explain differences in forage accumulation and canopy characteristics of five genotypes of *Brachiaria* spp. (Basilisk, Marandu, Mulato II, Piatã and Xaraés) in southeastern Brazil under three levels of nitrogen fertilization (220 kg of N ha^{-1} yr^{-1} in year 1 and 0 and 550 kg of N ha^{-1} yr^{-1} in year Year 3), and adapt the CROPGRO-Perennial Forage model (CROPGRO-PFM) to simulate the seasonal growth and production of these same genotypes, as well as to describe the adaptations and improvements made to the model. Both objectives were developed from an experiment of mechanically harvested plots with 15 cm stubble every 28 days and 42 days during the wet and dry season, respectively, from October 2013 to November 2014 (Year 1) and from September 2015 to September 2016 (Year 3). Response measurements included annual herbage accumulation, plant-part composition, canopy height, herbage bulk density, leaf area index, light interception, and tiller population density. The first objective showed us that there were differences between the genotypes, indicating great variability. Mulato II, Piatã and Xaraés were the most productive genotypes, producing an annual average of 12.1 Mg DM ha^{-1}. The plots fertilized with N produced 2.3 times more forage than the non-fertilized plots. The main contrast occurred between the Basilisk and Xaraés genotypes. Basilisk showed higher tiller population density (1512 vs. 854 tillers per m^2) and low leaf:stem ratio (2.4 vs. 12.8) in relation to Xaraés. While the second objective showed us that the model adaptation required the modification of parameters related to the partition between leaf and stem fractions, and the allocation intensity to shoot versus storage tissues based on photoperiod sensitivity. And after these modifications, the CROPGRO-PFM model performed well, providing realistic seasonal growth responses to seasonal climate variations and nitrogen fertilization. Morphological and structural characteristics of the canopy are valuable ways of understanding the management potential of different *Brachiaria* genotypes. And after adapting the model, we can apply it as a tool to simulate the growth and partitioning of the five *Brachiaria* genotypes in different scenarios.

Keywords: Tropical grasses; Forage production; Seasonal growth; Pastures; *Urochloa brizantha*; *Urochloa decumbens*; Crop models; DSSAT.

RESUMO

Brachiaria spp. desempenham um papel fundamental em sistemas de produção animal baseados em pastagens em regiões tropicais e subtropicais. A avaliação da produção de forragem e das características do dossel permitem entender as respostas da pastagem ao longo do tempo e auxiliam na identificação de genótipos mais produtivos com diferentes necessidades de manejo. Nossos objetivos foram comparar e explicar diferenças no acúmulo de forragem e características do dossel de cinco genótipos de *Brachiaria* spp. (Basilisk, Marandu, Mulato II, Piatã e Xaraés) no sudeste do Brasil cultivados com três níveis de adubação nitrogenada (220 kg de N ha^{-1} ano^{-1} no ano 1 e 0 e 550 kg de N ha^{-1} ano^{-1} no ano Ano 3), e adaptar o CROPGRO-Perennial Forage model (CROPGRO-PFM) para simular o crescimento sazonal e a produção desses mesmos genótipos, bem como descrever as adaptações e melhorias feitas no modelo. A partir de um experimento de parcelas colhidas mecanicamente com 15 cm de resíduo a cada 28 dias e 42 dias durante a estação chuvosa e seca, respectivamente, de outubro de 2013 a novembro de 2014 (Ano 1) e de setembro de 2015 a setembro de 2016 (Ano 3). Foram mensurados o acúmulo anual de forragem, composição de partes da planta, altura do dossel, densidade da forragem, índice de área foliar, interceptação de luz e densidade populacional de perfilhos. Observamos diferenças significativas entre os genótipos. Mulato II, Piatã e Xaraés foram os genótipos mais produtivos, produzindo uma média anual de 12,1 Mg MS ha^{-1}. As parcelas adubadas com N produziram 2,3 vezes mais forragem do que as parcelas não adubadas. O principal contraste ocorreu entre os genótipos Basilisk e Xaraés; O Basilisk apresentou maior densidade populacional de perfilhos (1512 vs. 854 perfilhos por m^2) e baixa relação folha:caule (2,4 vs. 12,8) em relação ao Xaraés. A adaptação do modelo exigiu a modificação de parâmetros relacionados à partição entre as frações de folha e caule, e a intensidade de alocação para parte aérea versus tecidos de armazenamento com base na sensibilidade ao fotoperíodo. E após essas modificações, o modelo CROPGRO-PFM teve um bom desempenho, fornecendo respostas realistas de crescimento sazonal às variações climáticas sazonais e fertilização com nitrogênio. As características morfológicas e estruturais do dossel são formas valiosas de entender o potencial de manejo de diferentes genótipos de *Brachiaria*. Após a adaptação do modelo, podemos aplicá-lo como ferramenta para simular o crescimento e particionamento dos cinco genótipos de *Brachiaria* em diferentes cenários.

Palavras-chave: Gramíneas tropicais; Produção de forragem; Crescimento sazonal; Pastagens; *Urochloa brizantha*; *Urochloa decumbens*; Modelos de colheita; DSSAT.

Inform Graphic.

Created by Daniel da Cunha and supervised by Márcio André Stefanelli Lara

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Agronomic traits and adaptation of the CROPGRO-Perennial Forage model to predict the growth of five *Brachiaria* genotypes

Objectives:

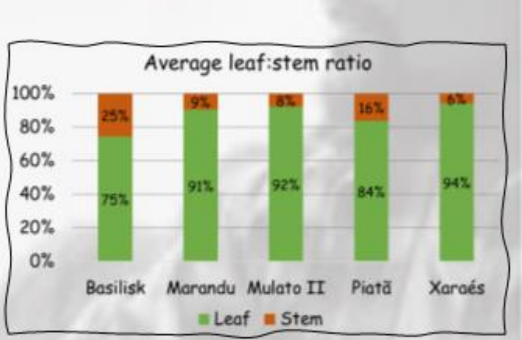
1. Compare and explain differences of five *Brachiaria* spp. *Basilisk, Marandu, Mulato II, Piatã and Xaraés*

2. Adapt the CROPGRO-Perennial Forage model to simulate the seasonal growth and production of the five genotypes

Agronomic comparison:

Nitrogen fertilization favors plant growth and canopy productivity

Brachiaria genotypes are different in structure and plant-part composition



Each genotype has different growth and requires different defoliation management

Model adaptation:

Soil water and N input parameters influence the simulation of crop variables

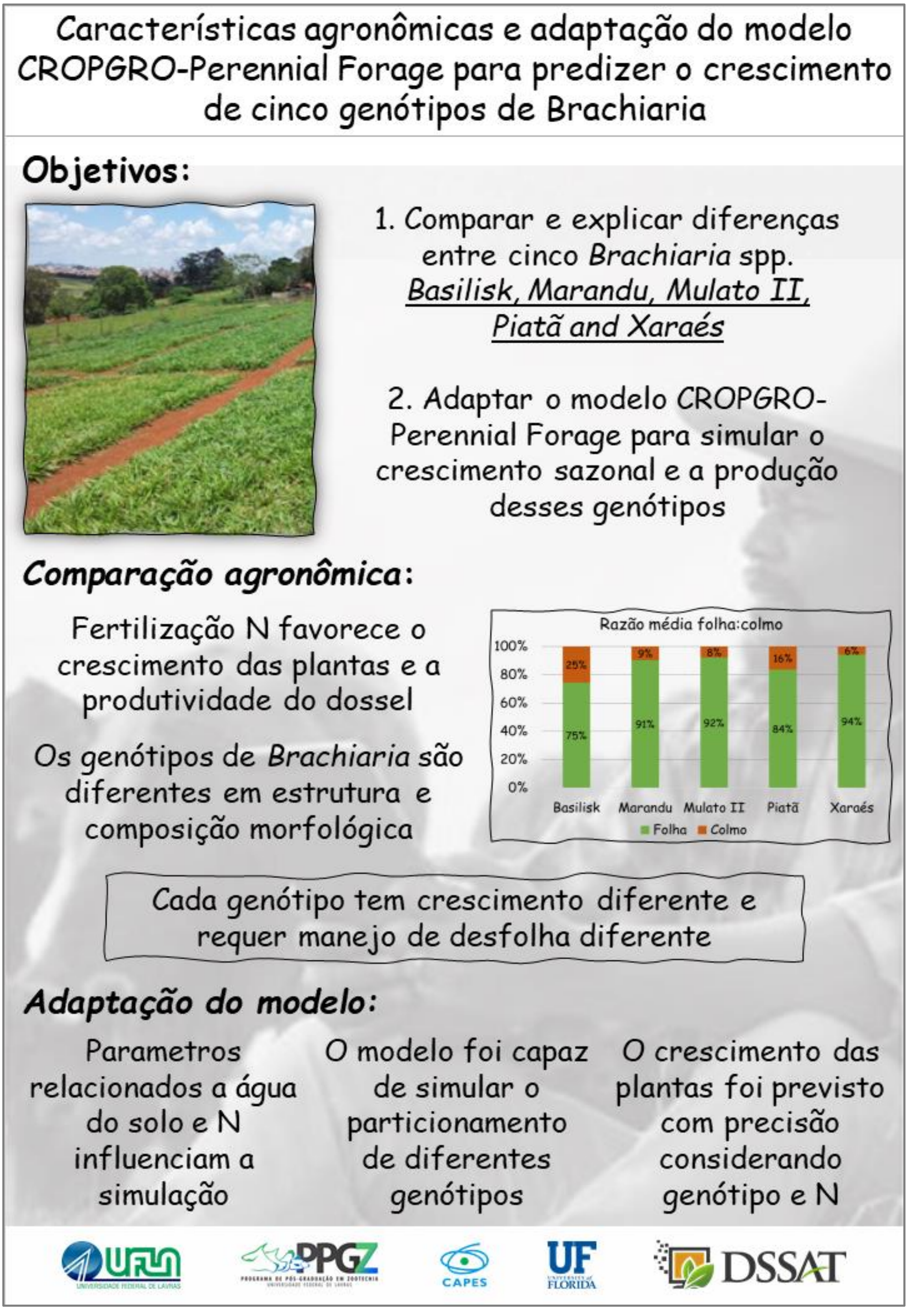
The model was able to simulate the partitioning of different *Brachiaria* genotypes

Plant growth was accurately predicted based on genotype and N input interaction

Doctoral Thesis in Animal Science at UFLA, defended on 02/25/2022.

Info Gráfico.

Criado por Daniel da Cunha e supervisionado por Márcio André Stefanelli Lara



Características agronômicas e adaptação do modelo CROPGRO-Perennial Forage para predizer o crescimento de cinco genótipos de Brachiaria

Objetivos:

1. Comparar e explicar diferenças entre cinco *Brachiaria* spp. *Basilisk, Marandu, Mulato II, Piatã and Xaraés*

2. Adaptar o modelo CROPGRO-Perennial Forage para simular o crescimento sazonal e a produção desses genótipos

Comparação agronômica:

Fertilização N favorece o crescimento das plantas e a produtividade do dossel

Os genótipos de *Brachiaria* são diferentes em estrutura e composição morfológica

Cada genótipo tem crescimento diferente e requer manejo de desfolha diferente

Adaptação do modelo:

Parametros relacionados a água do solo e N influenciam a simulação

O modelo foi capaz de simular o particionamento de diferentes genótipos

O crescimento das plantas foi previsto com precisão considerando genótipo e N

Tese de Doutorado em Zootecnia na UFLA, defendida em 25/02/2022.

SUMMARY

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FIRST SECTION

1. INTRODUCTION

The key to the success of forage-livestock systems is the efficient use of forage resources. This is directly related to the improvement, release and dissemination of new genotypes evaluated under different conditions, which present better productivity, adaptability and nutritional value of the forage resource. This is especially relevant in Brazil, where the world's largest commercial herd of cattle is mainly supported on native and cultivated pastures that occupy about 159 million hectares (ABIEC, 2020; IBGE, 2017).

In Brazil, grasses of the genus *Brachiaria* (syn. *Urochloa*), account for approximately half of the pasture area, occupying about 80 million hectares. While Marandu occupies about 50 million hectares, other cultivars, such as Xaraés, Mulato II and Piatã, are used as complementary alternatives (Jank *et al.*, 2014). Knowing that climatic factors such as rain, temperature and light influence the physiological and morphogenic processes of forage, promoting seasonality in forage production and its nutritional value (Reis *et al.*, 2013), many farmers adopt different grass genotypes, nitrogen fertilization and harvest management strategies to improve forage production and nutritional values over the year.

It is likely that for each of the pasture livestock production systems in Brazil there is a more suitable forage genotype and management strategies to be adopted that maximize the efficiency of the forage-livestock system. Therefore, it is important to work on the development and improvement of systems for predicting the potential of plant production that consider both agronomic and morphological aspects of plants. Through well-parameterized simulation tools and models, we can develop production scenarios combining the best cultivation, management and harvesting strategies to maximize livestock production efficiency.

In view of all that has been exposed, the first section of this research was dedicated to a bibliographic review of the main topics discussed here, while the second session was dedicated to the manuscripts generated. Manuscript number one is dedicated to the comparison and elucidation of the main differences related to herbage accumulation, morphological composition, and other agronomic traits of five different genotypes of *Brachiaria* under different levels of nitrogen fertilization. Manuscript number two is part of a series of studies that aim to adjust simulation models for forage grasses and is dedicated to adapting the CROPGRO-Perennial Forage model to predict the growth of the same five *Brachiaria* cultivars. And manuscript number three is focused on the development of a decision-making support system, aiming to predict the optimal harvest time of crops based on the concept of degree-days of development that was applied to predict the optimal harvest time of Marandu palisade grass after sowing, and can be adapted to different crops.

2. BACKGROUND

2.1. Forage grass of the genus *Brachiaria*

Brachiaria genotypes are warm-season perennial forages originated in tropical Africa and well adapted to tropical and subtropical areas of Brazil. They are considered fundamental for the development of the national beef industry due to the tolerance to acid and low fertility soils that prevail in the Brazilian forage-livestock systems (Lara *et al.*, 2021). As early as 1952, the genotype of *Brachiaria decumbens* (Stapf.) cv. Basilisk was one of the first forages to be introduced in Brazil, and it quickly spread throughout the national territory, due to its excellent adaptation to edaphoclimatic conditions (Sendulsky, 1978). Reports from 1975 already indicated that the cultivar had become a large monoculture, occupying extensive areas of the Brazilian savannas. Later, with the increase in the frequency of attacks by spittlebugs (*Deois* sp. and *Notozulia* sp.) and with the discovery of photosensitization in cattle, the demand for new forage options intensified. Even with the identification of limitations, Basilisk is still one of the most important *Brachiaria* cultivars in Brazil for grazing (Jank *et al.*, 2014; Pedreira *et al.*, 2017).

As an alternative to Basilisk, in 1984, the Brazilian Agricultural Research Corporation (EMBRAPA) release the *B. brizantha* (Hochst. Ex A. Rich.) Stapf. cv. Marandu with the differential of tolerating the attack of spittlebugs, reaching higher forage accumulation rates when compared to cultivar Basilisk (Nunes *et al.*, 1984). For these reasons, this genotype was widely recommended for planted pastures in Brazil, becoming one of the largest monocultures in the world, occupying about 50 million hectares, according to Jank *et al.* (2014). Despite its importance and dominance in the national territory, the recent “Marandu death syndrome” has contributed to the decline in productivity and persistence of this grass in central and northern Brazil. The causes of this syndrome today are related to high annual rainfall, poorly drained soils, and the presence of fungi in the soil (Junior *et al.*, 2016). This stimulated the search and development of forage alternatives to meet the demand and efficiency of the Brazilian forage-livestock systems.

As part of the development process new forage grass genotypes were released. In 2003, the cultivar Xaraés, in 2005, the cultivar Mulato II and, later, in 2007, the cultivar Piatã , were launched as an alternative to the other genotypes widely used in Brazil. *B. brizantha* (Hochst. Ex A. Rich.) Stapf. cv. Xaraés and Piatã were characterized by rapid growth and regrowth, productivity, and high herbage accumulation, both favoring the production of animals per area. While the cultivar Xaraés stands out for its good tolerance to spittlebugs and poorly drained soils, the cultivar Piatã stands out for its adaptability and rapid establishment when planted in medium fertility and well-drained soils, and for its high nutritional value (Euclides *et al.*, 2008, 2009). The *Brachiaria* cv. Mulato II, a three-way hybrid of *Brachiaria brizantha*, *Brachiaria ruziziensis* and *Brachiaria decumbens*, was a result of three generations of crossing and selection conducted by CIAT. The Mulatto II genotype stands out for its high quality and forage production, resistance to the main species of spittlebugs and moderate susceptibility to foliar fungi, in addition to showing good adaptation to acidic and well-drained tropical soils (Vendramini *et al.*, 2012).

2.2. Nitrogen fertilization on plant growth

Nitrogen (N) is the element most demanded mineral by plants and the element with the highest concentration in plant tissues. It constitutes proteins, nucleic acids, and other cellular components, in addition to participating in the composition of phytohormones (Cantarella, 2007). In general, N is available in the soil solution and is absorbed by the plant's root system and transported to the shoot through the xylem and easily distributed by the plant via the phloem. When N levels are at adequate levels, the color of the leaves tends to dark green, otherwise it causes chlorosis (yellowing) of the leaves (Vilanova *et al.*, 2018).

The positive effects of N are directly related to the greater accumulation and production of biomass of forage grasses, due to the increase in the rate of appearance and elongation of leaves (morphogenic characteristics), the increase in population density, the number of leaves per tiller and the leaf area rate (structural characteristics) (Silva *et al.*, 2016). At the physiological level, the effects of increased N availability for forage grasses are related to the promotion of photosynthetic activity, mobilization of photo-assimilates from reserves (C and N), mainly during the regrowth process. All these effects are modulated mainly by the genetic characteristics of the plants as well as their interaction with environmental, biotic, and abiotic factors, in addition to the level of N availability itself. Thus, contrasting levels of different nitrogen fertilization promote the dynamics of pasture growth.

The literature reports that the greater the availability of N in the forage grass production system, the greater the productivity and the greater the efficiency of forage harvest. The increase in N availability, in addition to accelerating the growth and development of forage plants, requires important management adjustments to ensure the maintenance of the nutritive value of the harvested forage. Fagundes *et al.* (2005) and Barcellos *et al.* (2011) reported that higher N doses induce rapid growth of *B. decumbens*, which requires the adoption of frequent grazing under intermittent stocking or adoption of lower management heights under continuous stocking (20 cm), under penalty of a drop in nutritional value due to the greater accumulation of stems (Pedreira *et al.*, 2017). Therefore, the correct understanding of the modulation of the effects of N fertilization in forage grasses constitutes an opportunity to maximize plant performance, increasing the production of leaves and biomass with high nutritional value and, at the same time, maximizing animal performance through adjustments in the harvest management of the forage produced.

2.3. Uses and application of crop models

Modeling comes from an area of knowledge called systems analysis, which aims to conceptualize, integrate, and generalize scientific knowledge through the simplification of complex systems. A mathematical model can be composed of an equation or a set of equations to represent biophysical processes (Rickert *et al.*, 2000). More specifically, mathematical models for simulating crop systems are useful: (i) in practice, to plan, assist in decision making and maximize the production efficiency of systems; (ii) in science, as tools to organize and identify gaps in knowledge about the physical and biological processes of plant growth and development, as well as their interrelationships with environmental variables; and (iii) in the simulation of current and future scenarios in the face of climate change.

Especially for forage-livestock systems, the intensification of production processes determines the efficiency and viability of the farms. Forage production simulation models are useful tools to project pasture management over time, considering regrowth cycles and accumulated forage use. Some models can even consider the growth of plants in the forage canopy, as well as the removal of forage by animals, and aspects related to nutrient cycling via litter, integrating the agronomic traits with animal production in a pasture ecosystem. In addition, these models make it possible to assess the influence of climate on productivity and seasonality of production over the years, considering current and future agricultural and environmental scenarios, in addition to measuring the possible impacts of climate change on production systems (Trnka *et al.*, 2006; Zhang *et al.*, 2006).

2.4. CROPGRO-Perennial Forage model

The adaptation of the CROPGRO perennial forage model began with Kelly (1995) simulating from *Paspalum Flügge*, a perennial grass with C4 metabolism, commonly used in crop rotation with peanuts and corn, in Florida, USA. Later, the model was adjusted to simulate *B. decumbens* cv. Basilisk, using data from the International Network for the Assessment of Tropical Pastures, CIAT, Colombia (Giraldo *et al.*, 1998; 2001).

Rymph *et al.* (2004), evaluating the growth model of *P. notatum* in simulations for hay production, found that dry matter production, especially in months with lower temperatures, was overestimated by the model. However, after calibrating and adjusting the model parameters, more accurate estimates of the seasonal growth of plants were obtained throughout the production cycles. Even so, the authors recommended the inclusion of variables related to the storage of reserve carbohydrates in plants.

Also in 2004, Rymph developed a perennial version of the P. notatum simulation model, adding a state variable for storing C and N reserves, along with principles for using these reserves during the regrowth process, after defoliation or the complete death of plants in vernalization events. The added parameters were directly related to the post-harvest conditions of the residues and their influence on the dynamics of partitioning and use of nutrient reserves during the cycles of biomass accumulation and regrowth management. Alderman (2008) adapted the model for the simulation of *Cynodon* ssp. and even improved the model code to optimize the simulation of forage harvest and the cycling dynamics of the soil organic matter.

The CROPGRO model code for perennial forages has been continuously improved and adapted to simulate the growth of several other tropical forage genotypes. More recently, the model was adapted to estimate and simulate the growth of different tropical grasses (Pedreira *et al.*, 2011; Lara *et al.*, 2012; Pequeno *et al.*, 2014; Pequeno *et al.*, 2017; Brunetti *et al.*, 2021), considering morphophysiological information from an established forage canopy, such as characteristics related to specific leaf area, photosynthesis, partitioning and nutrient composition.

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SECOND SECTION

ARTICLE 1

Agronomic traits of five *Brachiaria* genotypes under N-fertilization levels in Southeastern Brazil

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Core Ideas

- Nitrogen fertilization favors plant growth and canopy productivity.
- *Brachiaria* genotypes are different in structure and plant-part composition.
- Each genotype has different growth and requires different defoliation management.

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[†]Abbreviations: CH, canopy height; DM, dry matter; HA, herbage accumulation; HBD, herbage bulk density; HM, herbage mass; LAI, leaf area index; LI, light interception; TPD, tiller population density; TPDr, reproductive tiller population density.

Abstract

The evaluation of productivity and canopy traits of forages allows the understanding of aspects related to the responses of pastures over time, providing opportunities for the identification of high productivity genotypes and different needs for defoliation management. The aim of this study was to compare and explain differences in herbage accumulation and canopy traits of five *Brachiaria* spp. (Basilisk, Marandu, Mulato II, Piatã and Xaraés) in southeastern Brazil under three N-fertilization levels (220 kg of N ha^{-1} yr^{-1} in Year 1, and 0 and 550 kg of N ha^{-1} yr^{-1} in Year 3). Plots were mechanically harvested to 15-cm stubble every 28 d and 42 d during the rainy and dry season, from October 2013 to November 2014 (Year 1) and from September 2015 to September 2016 (Year 3). Response measurements included annual herbage accumulation, plant-part composition, canopy height, herbage bulk density, leaf area index, light interception, and tiller population density. There were differences between the genotypes for these traits, indicating great variability. Mulato II, Piatã and Xaraés were the most productive genotypes, producing an annual average of 12.1 Mg DM ha^{-1}. The N-fertilized plots produced 2.3 times more forage than the non-fertilized plots. The main contrast occurred between the Basilisk and Xaraés genotypes; Basilisk had higher tiller population density (1512 *vs.* 854 tillers per m^2), and low leaf:stem ratio (2.4 *vs.* 12.8) compared to Xaraés. This study showed that the morphological and structural characteristics of the canopy are valuable ways to understand the potential management of different *Brachiaria* genotypes.

Keywords: Forage production; Tiller population density; Seasonal growth; Pastures; *Urochloa brizantha*; *Urochloa decumbens*.

1. Introduction

Originally from the African continent, the forage genus *Brachiaria* is now widely distributed across Brazil and has become the most used grass in the forage-livestock system (Luna *et al.*, 2016). The *B. decumbens* (Stapf.) cv. Basilisk was one of the first forages to be introduced in Brazil, and it quickly spread throughout the country, due to its excellent adaptation to edaphoclimatic conditions (Sendulsky, 1978). However, with the increase in the frequency of pest attacks by spittlebugs and with the discovery of a response to photosensitization in cattle, alternative forage genotypes have been developed.

In 1984, the Brazilian Agricultural Research Corporation (EMBRAPA) released *B. brizantha* (Hochst. Ex A. Rich.) Stapf. cv. Marandu that has the advantage of tolerating spittlebug attack and producing a higher forage accumulation rate than other genotypes that have been previously released (Argel *et al.*, 2007). Recently, however, a phenomenon called “Marandu death syndrome” has contributed to the decline in productivity and persistence of this genotype in the north-central regions of Brazil (Junior *et al.*, 2016), which has triggered breeding programs for the release of new genotypes.

As part of the breeding process, EMBRAPA released the genotype Xaraés in 2003, the genotype Mulato II in 2005, and, later, the cultivar Piatã in 2007, as new alternatives. *B. brizantha* (Hochst. Ex A. Rich.) Stapf. cv. Xaraés and Piatã are characterized by rapid growth, and high herbage accumulation. Xaraés stands out for its good tolerance to spittlebugs and poorly drained soils, while Piatã stands out for its fast establishment and high nutritional value (Euclides *et al.*, 2008). Mulato II, a three-way hybrid of *B. brizantha*, *B. ruziziensis* and *B. decumbens*, was a result of three generations of crossing and selection conducted by CIAT and stands out for its high quality and herbage production, its resistance to the main species of spittlebugs and a moderate susceptibility to foliar fungi (Vendramini *et al.*, 2012).

There might be more suitable forage genotypes for adoption on individual farms in Brazil, with the objective of maximizing the efficiency of the livestock-forage systems. Thus, the objective of this study was to compare and explain differences in herbage accumulation and canopy traits of five *Brachiaria* genotypes under N-fertilization levels.

2. Materials and Methods

2.1 Research Site, Treatments, and Experimental Design

A field trial was conducted at the Federal University of Lavras in Lavras, state of Minas Gerais, Brazil (21°14' S, 44°58' W and 919 m above sea level). The region has a subtropical climate, classified as CWa, according to Köppen's method (Alvares *et al.*, 2013). The soil of the experimental area is classified as Dystrophic Red Latosol with clay texture. The chemical characterization of the soil showed pH of 6.2; organic matter of 40 g dm^{-3}; P of 2.0 mg dm^{-3}; K^+ of 75.0 mg dm^{-3}; Ca^{2+} of 3.1 $cmol_c dm^{-3}$; Mg^{2+} of 0.7 $cmol_c dm^{-3}$; and H + AL of 2.6 $cmol_c dm^{-3}$; sum of bases of 4.1 $cmol_c dm^{-3}$; cation exchange capacity of 6.6 $cmol_c dm^{-3}$; and base saturation of 61%. The proportion of clay was 549 g kg^{-1}, and silt was 112 g kg^{-1}. Daily weather data were obtained from a meteorological station that was located at 1.2 km from the experimental area (Table 1). The water balance and evapotranspiration was calculated using the method described by Thornthwaite and Mather (1955) (Figure 1).

The experimental design was a randomized complete block with four replications. Treatments were five *Brachiaria* genotypes including three [*Brachiaria brizantha* (Hochst. Ex A. Rich.) Stapf.] (Marandu, Piatã and Xaraés), one *Brachiaria decumbens* (Stapf.) (Basilisk), and a three-way hybrid of *B. brizantha*, *B. ruziziensis* and *B. decumbens* (Mulato II). Grasses were sown at a rate of 6 kg ha^{-1} viable seed on April 2012 in 8-m by 4-m plots, and harvested at 15-cm stubble height every 28 d during the rainy season and every 42 d during the dry season. The agronomic responses were measured for a period of two years from October 2013 to

November 2014 (Year 1) and from September 2015 to September 2016 (Year 3), totaling a total of 23 regrowth cycles.

In Year 1, nitrogen and potassium were split-applied using $(NH_4)_2SO_4$ and KCl as fertilizers after each harvest, to provide a total annual application of 220 kg N and K_2O ha^{-1}. From December 2014 to September 2015, the plots were harvested and fertilized with the equivalent of 220 kg N and K_2O ha^{-1} $year^{-1}$, but no data were collected. In Year 3, there were two levels of nitrogen fertilization, the non-fertilized treatment, equivalent to 0 kg of N ha^{-1} yr^{-1}, and the N-fertilized treatment, equivalent to 550 kg of N ha^{-1} yr^{-1}.

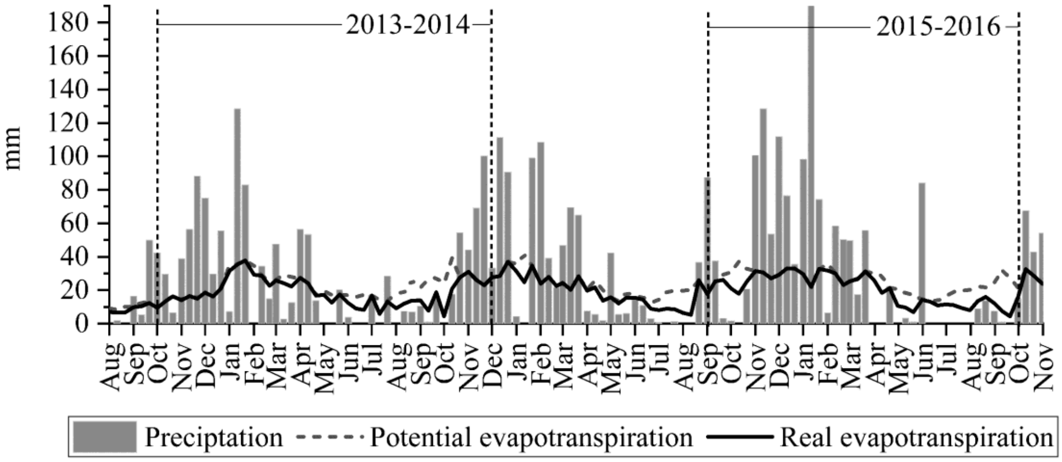

Figure 1. Water balance from August 2013 to November 2016 in Lavras, MG, Brazil (Potential and Real Evapotranspiration computed based on Thornthwaite and Mather, 1955). Dashed lines shows the experimental periods for Year 1, and Year 3.

Table 1. Monthly weather data during the experimental periods and 30-yr average in Lavras, Minas Gerais, Brazil.

Weather variable	Rainy season						Dry season					
	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.
2013-2014												
Avg. solar radiation, MJ m^{-2} d^{-1}	18.1	19.2	20.9	26.4	24.0	19.6	17.2	15.6	13.7	13.2	18.3	18.2
Max. temperature, °C	26.4	27.4	28.8	31.5	30.7	29.1	27.7	25.4	25.2	23.5	26.8	28.8
Min. temperature, °C	15.7	17.4	18.8	18.1	18.1	18.1	16.3	13.2	12.9	12.3	12.3	14.6
Total rainfall, mm	78	183	160	219	49	63	123	20	4	45	14	28
2014-2015												
Avg. solar radiation, MJ m^{-2} d^{-1}	22.4	19.7	21.4	24.9	20.6	16.2	17.1	13.2	13.3	13.2	18.2	15.8
Max. temperature, °C	29.8	28.3	29.5	31.9	28.5	27.6	27.4	24.1	24.0	25.3	27.3	28.1
Min. temperature, °C	16.0	17.8	18.3	18.9	18.7	18.3	16.9	14.5	13.1	13.1	12.9	16.5
Total rainfall, mm	72	214	235	103	169	181	15	54	28	2	37	128
2015-2016												
Avg. solar radiation, MJ m^{-2} d^{-1}	22.2	18.8	21.0	18.8	22.0	17.7	19.0	13.8	13.4	15.9	17.4	20.2
Max. temperature, °C	31.3	29.7	29.8	28.3	30.5	29.3	29.4	25.7	23.4	25.8	27.3	29.9
Min. temperature, °C	18.3	19.1	19.1	18.9	19.4	18.9	17.1	14.6	12.3	11.7	13.0	15.4
Total rainfall, mm	22	283	224	401	127	111	22	5	84	0	30	1
1981-2010 †												
Avg. day length (h d^{-1})	12.5	13.0	13.4	13.3	12.8	12.2	11.6	11.0	10.6	10.7	11.1	11.8
Max. temperature, °C	28.1	27.9	27.9	28.5	29.3	28.6	27.6	25.2	24.5	24.8	26.7	27.6
Min. temperature, °C	16.2	17.2	18.0	18.3	18.3	17.8	16.1	13.3	11.6	11.1	12.1	14.3
Total rainfall, mm	115	188	291	273	210	168	57	46	20	11	13	71

† Average monthly historical weather (1981-2010).

2.2 Response Measurements

2.2.1 Annual and Seasonal Herbage Accumulation, and Plant-Part Composition

At the end of each regrowth cycle, the forage inside two 0.5-m^2 quadrats per plot was clipped to a 15-cm stubble height and weighed for fresh weight. From each herbage mass (HM) sample, two subsamples of approximately 600 g was taken. One subsample was used to determine the dry matter concentration (DM), while the other was used to determine the morphological composition of the plants, separating them into green leaf (leaf blade), stem (stem + leaf sheath) and dead material. The subsamples were dried in a forced ventilation oven at 55ºC for 72 h to calculate the DM. The annual herbage accumulation (HA) was obtained by summing HM across all regrowth cycles.

After HM samples were taken, the entire plot was mechanically harvested at 15-cm stubble height and fertilized. Each year was divided into a rainy season (21 September to 20 March) and dry season (21 March to 20 September).

2.2.2 Canopy Height, Herbage Bulk Density, Leaf Area Index, and Light Interception

Pre-harvest canopy height (CH) was measured on the last day of each regrowth cycle (immediately before plot harvest) by taking 24 readings using a light polyethylene sheet and a ruler. Herbage bulk density (HBD) of the harvested stratum was calculated by dividing the harvested HM by the mean non-extended canopy height (CH) minus the stubble height.

The leaf area index (LAI) and light interception (LI) at pre-harvest were measured every cycle using a LAI-2200 canopy analyzer (LI-COR, Lincoln, NE, USA) according to Welles and Norman (1991). Measurements of canopy condition were conducted at representative sites (by visual assessment) by taking eight readings at the bottom of the canopy, as close as possible (~ 2 cm) to the surface of the soil, and two above the canopy for reference calibration.

2.2.3 Tiller Population Density

Tiller population density was measured on the last day of the regrowth for only two cycles, each representative of the rainy and dry seasons. The measurements were performed by counting the total number of tillers (TPD), and the reproductive tillers (TPDr) contained in two rectangles of 0.10-m^2 per plot.

2.3 Statistical Analysis

Data were analyzed with PROC MIXED of SAS (SAS Institute, Cary, NC, USA) with repeated measures and using the restricted maximum likelihood (REML) method, for each experimental period (Year 1 and Year 3). For Year 1, the genotype, season, and their interaction were considered fixed effects, while for Year 3, the genotype, N-fertilization levels, season, and their interactions were considered fixed effects. In both analyses, the replication (block) and interactions involving blocks were considered random effects. Seasons (rainy vs. dry) were analyzed as repeated measures. The choice of the covariance matrix was made using the Akaike information criterion (AIC) (Wolfinger, 1993). Genotypes were compared using Tukey's test ($P < 0.05$), while seasons and N-fertilized and non-fertilized treatments were compared using PDIFF by Student's t test ($P < 0.05$).

3. Results and Discussion

3.1 Annual and Seasonal Herbage Accumulation

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the annual HA was different ($P < 0.05$) among the five genotypes evaluated. The most productive genotype was Xaraés, with 32% (3.1 Mg DM ha^{-1}) more herbage when compared to Marandu, the least productive genotype. On average, Mulato II, Piatã and Xaraés accumulated about 16% (1.7 Mg DM ha^{-1}) more herbage when compared to the average accumulated by Basilisk and Marandu (Table 2). This result shows that forage breeding programs are evolving and generating new and more productive alternative genotypes.

For Year 3, the annual HA was not influenced by the G × N interaction (P = 0.5766). However, there was an effect of N (P < 0.0001), indicating that the N-fertilized plots accumulated about 2.3 times (7.0 Mg DM ha^{-1}) more herbage when compared to the non-fertilized plots (Table 2). However, there were no differences among the genotypes within the N-fertilized and non-fertilized plots in Year 3, thus indicating that N availability was a more important factor for maximizing annual HA.

Regardless of the levels of N-fertilization evaluated, variations in HA resulted from the interaction between the environment and plant genetics. Comparing the environmental conditions between Year 1 and Year 3, we observed less daily total solar radiation (19.0 vs. 18.2 MJ m^{-2} d^{-1}), an increase of the minimum temperature (15.8 vs. 16.5°C), an increase in precipitation (1272 vs. 1438 mm) and an increase in the accumulated water deficit (160.0 vs. 199.9 mm) in Year 3 (Table 1 and Figure 1). Therefore, the integrated understanding of these factors and their effects on plant growth, can help the development of pasture management strategies, considering the requirements for each genotype.

Table 2. Annual herbage accumulation (Mg DM ha^{-1}) of five *Brachiaria* genotypes as affected by the genotype × nitrogen interaction over different experimental periods and treatments in Lavras, MG, Brazil.

Year	Nitrogen (N)	Genotype (G)					SEM	Mean	*P value**
		Basilisk	Marandu	Mulato II	Piatã	Xaraés			
1	220	11.0 bc	9.7 c	11.4 abc	12.2 ab	12.8 a	0.54	11.4	-
3	0	3.7	2.5	2.6	2.7	3.3	0.89	3.0	0.5766
	550	11.6	9.3	9.6	8.5	10.8		10.0	
	P value$^+$							<.0001	

* *P value* of the G × N interaction (Year 3); $^+$ *P value* of the N effect (Year 3); Means followed by the same letters in the lines are considered statistically similar by the Tukey test (P < 0.05).

Separating the annual HA for the rainy and dry season of Year 1 and Year 3, we obtained the seasonal pattern of HA for each genotype. The HA was influenced by the G × N interaction for Year 1 (P = 0.0023), and Year 3 (P < 0.0001) (Table 3). However, for Year 3, there was no difference (P > 0.05) for HA between seasons comparing N-fertilized and non-fertilized genotypes (data not shown).

For both Year 1 and Year 3, Basilisk had the highest proportion of HA in the rainy season, an average of 80% among the years. For Year 1, Basilisk accumulated 9% more herbage when compared to Xaraés, and for Year 3, it accumulated 18% more herbage when compared to Mulato II. On average, the proportion of HA in the rainy season represented about 74% of the annual HA, averaged over genotypes and the N-fertilization levels (Table 3). These results are consistent with other reports for the Central region of Brazil, in which 75 to 85% of the annual biomass production occurs during the warm-rainy season (Barioni *et al.*, 2007).

For the two years that were evaluated in this study, the five genotypes showed a greater HA during the rainy season ($P < 0.0001$). Environmental conditions during the warm-rainy season, such as longer photoperiod, higher temperature and higher rainfall, favor forage growth regardless of N availability. The recently released genotypes, i.e., Mulato II, Piatã and Xaraés, had a lower HA seasonality. Among them, the Mulato II genotype stood out for a better distribution of annual HA during the rainy and dry seasons for the years that were evaluated, regardless of the N-fertilization level (Table 3).

However, the total HA throughout the year, as well as its seasonal distribution, do not fully explain the differences of each genotype. Therefore, a study of the plant-part composition is helpful for the differentiation of the evaluated grasses.

Table 3. Proportion herbage accumulation (%) of five *Brachiaria* genotypes in rainy versus dry season as affected by the genotype × season interaction over different experimental periods and treatments in Lavras, MG, Brazil.

Year	Season (S)	Genotype (G)					SEM	Mean	*P value*[*]
		Basilisk	Marandu	Mulato II	Piatã	Xaraés			
1	Rainy	82.1 a	77.0 ab	77.1 ab	75.8 b	75.1 b	1.46	77.4	0.0023
	Dry	17.9 b	23.0 ab	22.9 ab	24.2 a	24.9 a		22.6	
	P value[++]			<.0001					
3	Rainy	77.6 a	68.0 bc	65.6 c	70.2 bc	71.9 ab	2.11	70.7	<0.0001
	Dry	22.4 c	32.1 ab	34.4 a	29.8 ab	28.2 bc		29.3	
	P value[++]			<.0001					

[*] *P value* of the G × S interaction (Year 1, and Year 3); [++] *P value* of the effect of S within G (Year 1, and Year 3); Means followed by the same letters in the lines are considered statistically similar by the Tukey test ($P < 0.05$).

3.2 Annual Plant-Part Composition

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the annual accumulation of leaves and stems was different ($P < 0.05$) among the five genotypes that were evaluated. Xaraés accumulated 41% (3.6 Mg DM ha^{-1}) more leaf mass and 82% (1.8 Mg DM ha^{-1}) less stem mass when compared to Basilisk (Table 4).

For Year 3, the annual leaf accumulation was not influenced by the G × N interaction ($P = 0.5138$). However, there was an effect of N ($P <0.001$), indicating that the N-fertilized plots accumulated about 2.6 times (5.9 Mg DM ha^{-1}) more leaf mass when compared to the non-fertilized plots (Table 4). Considering the average of the N-fertilized and non-fertilized genotypes, Xaraés had 49% (2.1 Mg DM ha^{-1}) higher annual leaf accumulation when compared to Piatã. The annual accumulation of stems was influenced by the G × N interaction ($P = 0.0005$), indicating that the N-fertilized plots accumulated about 1.7 times (0.94 Mg DM ha^{-1}) more stems when compared to the non-fertilized plots. Considering the average of the genotypes N-fertilized and non-fertilized, Basilisk had a higher annual stem accumulation, about 3.4 times (1.7 Mg DM ha^{-1}) greater when compared to Xaraés (Table 4).

Even with the differences imposed by the N-fertilized and non-fertilized treatments, the genotypes kept the proportion of accumulation of leaves and stems constant. The increase in N availability did not change the proportion of the morphological components of the five genotypes. For Year 3, considering the average of N-fertilized and non-fertilized genotypes, Xaraés had a leaf:stem ratio that was 4.3 times greater than Basilisk (leaf:stem of 12.8 vs. 2.4). Lara *et al.* (2021) verified that the leaf:stem ratio of different genotypes of *Brachiaria* spp. remained as a constant genetic trait, based on evaluation of the growth of plants subjected to different levels of N-fertilization, frequency, and intensity of defoliation. These results reaffirm that a grazing management strategy based on genetic traits such as the number of live leaves per tiller, leaf:stem ratio and canopy leaf area index is a good strategy, as these traits are stable regardless of environmental factors (Fulkerson and Donaghy, 2001).

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the annual accumulation of dead material was not different ($P > 0.05$) among the five genotypes that were evaluated, on average 0.13 Mg DM ha^{-1} (Table 4). For Year 3, the annual accumulation of dead material was not influenced by the G × N interaction ($P = 0.8804$). However, there was an effect of N ($P < 0.001$), indicating that the N-fertilized plots accumulated about 1.8 (0.24 Mg DM ha^{-1}) times more dead material when compared to the non-fertilized plots. According to Lemaire *et al.* (2009), N-fertilization accelerates plant growth and with it the senescence rate. On average, among the genotypes for both the N-fertilized and non-fertilized treatments, Basilisk had 72% (0.13 Mg DM ha^{-1}) more dead material when compared to Xaraés (Table 4).

The association of total and seasonal HA throughout the year, and the plant-part composition, still does not account for all the differences among the evaluated genotypes. According to Macedo *et al.* (2021), understanding the spatial distribution of the morphological components and their influence on structural characteristics and canopy light interception can help with defining pasture management strategies according to the specificities of each genotype.

Table 4. Annual leaf, stem, and dead material accumulation (Mg DM ha^{-1}) of five *Brachiaria* genotypes as affected by the genotype × nitrogen interaction over different experimental periods and treatments in Lavras, MG, Brazil.

Year	Nitrogen (N)	Genotype (G)					SEM	Mean	*P value**
		Basilisk	Marandu	Mulato II	Piatã	Xaraés			
---------- *Leaf* ----------									
1	220	8.7 c	9.0 c	10.7 b	10.6 b	12.3 a	0.54	10.3	-
3	0	2.4	2.1	2.2	1.7	3.0	0.37	2.3	0.5138
	550	7.9	8.0	8.4	6.8	9.8	0.86	8.2	
	Mean	5.2 ab	5.1 ab	5.3 ab	4.3 b	6.4 a	0.55		
	P value^{+}							<.0001	
---------- *Stem* ----------									
1	220	2.2 a	0.5 c	0.6 c	1.5 b	0.4 c	0.13	1.04	-
3	0	1.0 a	0.4 ab	0.3 ab	0.8 ab	0.3 b	0.08	0.56	0.0005
	550	3.3 a	1.1 b	0.9 b	1.4 b	0.8 b	0.23	1.50	
	Mean	2.2	0.7	0.6	1.1	0.5	0.13		
	P value^{++}	<.0001	0.0109	0.0307	0.0236	0.0425			
---------- *Dead material* ----------									
1	220	0.11	0.13	0.10	0.16	0.15	0.023	0.13	-
3	0	0.17	0.13	0.14	0.14	0.08	0.015	0.13	0.8804
	550	0.44	0.38	0.41	0.33	0.28	0.067	0.37	
	Mean	0.31 a	0.26 ab	0.28 ab	0.24 ab	0.18 b	0.033		
	P value^{+}							<.0001	

* *P value* of the G × N interaction (Year 3); $^{+}$ *P value* of the N effect (Year 3); $^{++}$ *P value* of the effect of N within G (Year 3); Means followed by the same letters in the lines are considered statistically similar by the Tukey test ($P < 0.05$).

3.3 Pre-harvest Canopy Height, Herbage Bulk Density, Leaf Area Index, and Light Interception

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the CH was different ($P < 0.05$) among the five genotypes that were evaluated. Xaraés was about 20% (4.4 cm) taller when compared to Marandu (Table 5).

For Year 3, CH was not influenced by the G × N interaction ($P = 0.7315$). However, there was an effect of N ($P < 0.001$), indicating that the N-fertilized plots grew about 24% (4.1 cm) taller when compared to the non-fertilized plots (Table 5). The increase in the availability of N also influences canopy traits such as CH, stem elongation and tillering dynamics (Silva *et al.*, 2016). Considering the average of the N-fertilized and non-fertilized genotypes, Xaraés was 10% (1.9 cm) taller compared to Piatã (Table 5).

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the HBD was different (P < 0.05) among the five genotypes that were evaluated (Table 5). Marandu, Mulato II and Piatã were 14% (15.1 Kg DM ha^{-1}cm^{-1}) denser than Xaraés when the plots were clipped at a stubble height of 15 cm.

For Year 3, the HBD was not influenced by the G × N interaction (P = 0.2320). However, there was an effect of N on Piatã (P = 0.0161). For Piatã, the non-fertilized plots were about 52% (70.4 Kg DM ha^{-1}cm^{-1}) denser when compared to the N-fertilized plots (Table 5). This genotype, when not fertilized, maintained the accumulated herbage closer to the ground. This can be explained by the increase in the tiller population density and the greater participation of leaves and stems in this portion clipped from the canopy above the 15-cm clipping height (Fagundes *et al.*, 2006).

Considering the average of the genotypes for both N-fertilized and non-fertilized, Basilisk and Piatã had an average HBD that was about 28% (38 Kg DM ha^{-1}cm^{-1}) denser than the other genotypes (Table 5). Only for Piata, the increase in N availability influenced the elongation of the stem and CH, reducing the HBD. We can also highlight that the N-fertilization of Mulato II and Xaraés had a slight influence on the HBD, despite the increase in the proportion of leaves in the HM. For these genotypes, the addition of N favors the opportunity to select leaves for grazing animals. According to Newman *et al.* (2002), it is possible to increase animal production as the proportion of leaves increases while the HBD is maintained.

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the LAI and a LI were not different (P > 0.05) among the five genotypes. On average, the LAI was 1.7 m^{2}m^{-2} and a LI was 72.1% when the plots were clipped at a stubble height of 15 cm (Table 5).

For Year 3, there was no effect of the G × N interaction on LAI (P = 0.8451) and LI (P = 0.8843). However, there was an effect of N (P < 0.0001) for both traits. The N-fertilized plots had an LAI that was about 24% higher (0.4 m^{2}m^{-2}) and an LI that was about 11% higher (7.5%), when compared to the non-fertilized plots (Table 5). This indicates that LAI and LI are highly correlated and influenced by N-fertilization (Yasuoka *et al.*, 2018). Considering the average of the genotypes for both the N-fertilized and non-fertilized treatments, Mulato II showed an increase of 11% (0.2 m^2m^{-2}) in LAI and 5% (3.4%) in LI when compared to the Piatã (Table 5).

For Year 3, the N-fertilized plots had a lower HBD. The increase in the availability of N favors the increase in LI and at the same time causes a decrease in CH. This is supported by the trade-off dynamics between tiller size and tiller population density in pastures under the same harvest management. According to Macedo *et al.* (2021), the increases in LI and HBD between regrowth cycles occurs due to the increase in the tiller population density, or even due to the reduction in leaf angle of the plants. Macedo *et al.* (2021) also state that the tiller population density can be used as a grazing management tool because it combines the effects of LI with CH. Therefore, the study of tillering dynamics over time becomes relevant, since it associates important canopy variables, such as CH, LI, HBD, and tillering during regrowth cycles. The understanding of these dynamics will provide more support for the development of refined pasture management strategies for each genotype that was evaluated in this study.

Table 5. Pre-harvest canopy height, herbage bulk density, leaf area index, and light interception of five *Brachiaria* genotypes as affected by the genotype × nitrogen interaction for different experimental periods and treatments in Lavras, MG, Brazil.

Year	Nitrogen (N)	Genotype (G)					SEM	Mean	*P value**
		Basilisk	Marandu	Mulato II	Piatã	Xaraés			
------------------------------------ *Canopy height (cm)* ------------------------------------									
1	220	24.2 b	22.3 b	22.7 b	24.1 b	26.7 a	0.48	24.0	-
3	0	17.0	16.5	16.8	16.1	17.7	0.51	16.8	0.7315
	550	20.6	20.5	20.7	20.2	22.4	0.46	20.9	
	Mean	18.8 b	18.5 b	18.8 b	18.2 b	20.1 a	0.39		
	P value^{+}							<.0001	
--------------------------- *Herbage bulk density (Kg DM ha^{-1}cm^{-1})* ---------------------------									
1	220	110.3 bc	120.3 abc	133.4 a	122.8 ab	100.4 c	4.60	117.4	-
3	0	176.8	163.7	122.5	206.5	117.7	8.60	157.4	0.2320
	550	173.5	141.2	139.7	136.1	124.4		143.0	
	Mean	175.2	152.5	131.1	171.3	121.1	14.22		
	P value^{++}	0.8988	0.3997	0.5173	0.0161	0.7985			
---------------------------------- *Leaf area index (m^{2}m^{-2})* ----------------------------------									
1	220	1.7	1.7	1.8	1.6	1.7	0.07	1.7	-
3	0	1.7	1.6	1.8	1.6	1.6	0.06	1.7	0.8451
	550	2.1	2.1	2.2	2.0	2.1		2.1	
	Mean	1.9 ab	1.9 ab	2.0 a	1.8 b	1.9 ab	0.05		
	P value^{+}							<.0001	
----------------------------------- *Light interception (%)* ------------------------------------									
1	220	72.3	71.7	74.3	70.4	71.6	1.36	72.1	-
3	0	71.3	71.3	73.8	69.8	70.9	1.07	71.4	0.8843
	550	78.5	78.8	80.5	77.8	79.0		78.9	
	Mean	74.9 ab	75.1 ab	77.2 a	73.8 b	75.0 ab	0.91		
	P value^{+}							<.0001	

[*] *P value* of the G × N interaction (Year 3); [+] *P value* of the N effect (Year 3); [++] *P value* of the effect of N within G (Year 3); Means followed by the same letters in the lines are considered statistically similar by the Tukey test ($P < 0.05$).

3.4 Pre-harvest Tiller Population Density

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the TPD was different ($P < 0.05$) among the five genotypes. On average, Basilisk had 63% (594) more tillers per m^{2} when compared to the average of Piatã and Xaraés (Table 6). The reduced TPD of recently released genotypes, such as Piatã and Xaraés, indicates that breeding programs are developing more productive plants with morphological and architectural structures that result in a lower tiller density but larger tillers. Currently, breeding programs aim to maximize forage production by reducing the effects of seasonality and improving plant morphological composition and canopy structure at the tillering level.

For Year 3, the TPD was influenced by the G × N interaction ($P < 0.0022$), indicating that the N-fertilized plots had 49% more tillers per m^2 when compared to the non-fertilized plots (Table 6). The increase in N availability stimulates tillering, regardless of the genotype evaluated. There was an effect of N for Basilisk ($P < 0.0001$), Marandu ($P = 0.0001$) and Piatã ($P < 0.0001$), specifically indicating that for these genotypes, the N-fertilization resulted in 65% more tillers per m^2 when compared to the same non-fertilized genotypes (Table 6).

Considering the average over N-fertilized and non-fertilized treatments, Basilisk had 90% more tillers per m^2 than Xaraés (Table 6). The TPD patterns of Piatã and Basilisk genotypes were similar. Despite being a recently released genotype, Piatã has a dynamic tillering pattern with high variability throughout the year, and according to Giacomini *et al.* (2014), the high variability of tillering over time makes managing the forage production and harvesting system a challenge. Therefore, understanding the tillering pattern of genotypes throughout the year can help us to develop more efficient strategies for pasture management.

For Year 1, under a common N-fertilization level (220 kg of N ha^{-1}), the TPDr was different ($P < 0.05$) among the five genotypes evaluated. Basilisk had 3.5 times (21) more reproductive tillers per m^2 when compared with the average of the Marandu, Mulato II, Piatã and Xaraés (Table 6).

For Year 3, the TPDr was not influenced by the G × N interaction ($P = 0.3334$). However, there was an effect of N ($P < 0.0148$), indicating that the N-fertilized plots had about 53% (10) more reproductive tillers per m^2 when compared to the non-fertilized plots (Table 6), although the proportion of reproductive tillers to total tiller number was not different. The N-fertilization increases the TPDr due to accelerated growth for the same clipping frequency (McKenzie, 1998). Considering the average of the N-fertilized and non-fertilized genotypes, Basilisk had 1.5 time (21) more reproductive tillers per m^2 when compared to Xaraés (Table 6).

Comparing Basilisk to Marandu, Mulato II, Piatã and Xaraés, regardless of the level of N-fertilization, Basilisk had 3.5 times more reproductive tillers per m^2 than the others. This result can be explained by the Basilisk indeterminate flowering pattern. According to Gitari and Njarui (2016), flowering duration is negatively associated with the proportion of leaves, and consequently with herbage production. As one of the most important traits selected for the improvement of forage grasses, genotypes that have flowering concentrated at a certain time of the year have superior potential for use in forage-livestock systems. Plants that remain in the vegetative development stage for longer have less variation in morphological and nutritional composition.

Table 6. Annual pre-harvest tiller population density of five *Brachiaria* genotypes as affected by the genotype × nitrogen interaction for different experimental periods and treatments in Lavras, MG, Brazil.

Year	Nitrogen (N)	Genotype (G)					SEM	Mean	*P value**
		Basilisk	Marandu	Mulato II	Piatã	Xaraés			
	---------------------------- *Total tiller density (tillers/m²)* ----------------------------								
1	220	1540 a	1044 bc	1177 b	967 c	925 c	57	1130	-
3	0	1115 a	783 b	925 a	669 b	684 b	31	835	0.0022
	550	1852 a	1225 b	1108 b	1148 b	881 c	31	1243	
	Mean	1483	1016	1004	908	782	48		
	P value^{++}	<.0001	0.0001	0.0726	<.0001	0.0539			
	---------------------------- *Reproductive tillers (tillers/m²)* ----------------------------								
1	220	27 a	10 b	3 b	3 b	8 b	4	10	-
3	0	36	24	12	15	7	8	19	0.3334
	550	34	34	35	27	14	7	29	
	Mean	35 a	29 a	24 ab	21 ab	14 b	8		
	P value^{+}							0.0148	

* *P value* of the G × N interaction (Year 3); $^{+}$ *P value* of the N effect (Year 3); $^{++}$ *P value* of the effect of N within G (Year 3); Means followed by the same letters in the lines are considered statistically similar by the Tukey test ($P < 0.05$).

With the main aspects related to annual and seasonal HA (Tables 2 and 3) resulting from the combination of morphological plant traits (Table 4) and structural characteristics of the pasture (Table 5) and TPD (Table 6), we can generally infer the main differences among the five genotypes that were evaluated in this study. Therefore, we conclude that the main contrast between the five genotypes is between Basilisk and Xaraés.

Being an older released genotype, Basilisk is the pioneer and stands out for its high TPD, mainly TPDr, with greater participation of stems in the total HM accumulated above the stubble height and high HBD. By contrast, Xaraés stands out due to the lower TPD observed in the different evaluation conditions and lesser participation of the stems in the HM accumulated above the stubble height. These results show us that the morphological and structural composition of the plants has a direct effect on pasture productivity, and finally, on the efficiency of forage-livestock systems under farm conditions.

4. Conclusions

Considering all the observed response variables related to genotypes and N-fertilization levels, it is evident that each genotype must be treated independently. The recommendation to adopt one genotype over another must be strategic and its management must consider all aspects related to the persistence and stability of the canopy over time. Regardless of the N-fertilization level, the choice of genotypes by farmers should prioritize the recent releases such as Xaraés due to the high productivity, better distribution of annual HA, higher leaf:stem ratio, and lower TPD, when compared to older releases such as Basilisk. Each genotype has a distinct pattern of growth and, therefore, the requirements for defoliation management must be aligned with the objectives of the forage-livestock systems.

Author Contributions

Conceptualization, M.L., K.B. and G.H.; methodology and supervision, M.L.; investigation and data curation, D.C. and L.S.; formal analysis and writing – original draft, D.C. and S.S.; writing – review and editing, all authors. All authors have read and agreed to the published version of the manuscript.

Declaration of competing interest

The authors declare that there is no conflict of interest.

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SECOND SECTION

ARTICLE 2

Adapting the CROPGRO-Perennial Forage model to predict growth of five *Brachiaria* genotypes under N-fertilization

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Highlights

- CROPGRO-Perennial Forage model was calibrated for five *Brachiaria* genotypes.
- Soil water and N input parameters influence the simulation of crop variables.
- The model was able to simulate the partitioning of different *Brachiaria* genotypes.
- Plant growth was accurately predicted based on genotype and N input interaction.

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Abstract

Brachiaria spp. play a fundamental role in animal production for pasture-based systems in tropical and subtropical regions of the world. Despite the importance of *Brachiaria*, current perennial forage simulation models have not been evaluated for their ability to simulate the growth and production of different genotypes within a single species. The objectives of this research were, therefore, to evaluate and adapt the CROPGRO-Perennial Forage model (CROPGRO-PFM) for simulating the seasonal growth and production of five different *Brachiaria* genotypes, and to describe the adaptations and improvements made in the model. Growth analysis data for five genotypes, grown with different levels of N fertilization under rainfed conditions, were used to evaluate the model. The adaptation of the model was conducted in phases, comparing the simulations with measured data during each phase. The parameters that were modified include the partitioning between leaf and stem fractions, and the intensity of allocation to shoot versus storage tissues based on sensitivity to photoperiod. After these modifications, the CROPGRO-PFM model performed well, providing realistic responses of seasonal growth to seasonal weather variation and N fertilization. Therefore, the model can be applied as a tool to simulate the growth of *Brachiaria*, particularly to specify differences among genotypes in partitioning characteristics.

Keywords: Tropical grasses; Pasture; Herbage production; Crop models; Seasonal growth; DSSAT.

1. Introduction

Grasses of the genus *Brachiaria* originating from the African continent are well adapted to tropical and subtropical areas of Brazil and have become the most used grasses in national forage-livestock systems (Luna *et al.*, 2016). Even with the good adaptability of pioneer *Brachiaria* genotypes, forage grass breeding programs have been active during the recent decades to release more promising genotypes.

With the objective of helping farmers to also evaluate the best choice of genotype to be adopted, forage crop models based on mechanistic principles have been developed and improved considering different scenarios. The adaptation of the CROPGRO-Perennial Forage model (CROPGRO-PFM), for example, was initiated by Rymph (2004). CROPGRO-PFM simulates changes in plant-part composition and growth based on physiological processes, meteorological data, soil conditions and plant management (Rymph, 2004; Hoogenboom *et al.*, 2019). Later, several authors have contributed to the development of the CROPGRO-PFM model for the simulation of different forage grasses (Rymph *et al.*, 2004; Alderman *et al.*, 2008; Pedreira *et al.*, 2011; Lara *et al.*, 2012; Pequeno *et al.*, 2014; Pequeno *et al.*, 2017; Brunetti *et al.*, 2021).

So far, the model has been adapted to simulate the growth of “Marandu” palisadegrass, “Tanzania” guineagrass, “Tifton 85” bermudagrass, and “Mulato II” brachiariagrass. CROPGRO-PFM considers the morpho-physiological information of an established canopy, as well as characteristics related to the specific leaf area, photosynthesis, partitioning, and nutrient composition. Thus, the objective of this study was to evaluate the CROPGRO-PFM model for simulating the growth of five *Brachiaria* genotypes under different N-fertilization levels. Our null hypothesis was that the standard model developed for Marandu palisadegrass, can accurately simulate growth and forage production of other *Brachiaria* genotypes, requiring only parameterization of specific genotypic characteristics related to the cultivar and ecotype parameters.

2. Materials and Methods

2.1. Field experiment

The data used for model calibration and evaluation were collected in a field trial conducted at the Federal University of Lavras in Lavras, state of Minas Gerais, Brazil (21°14' S, 44°58' W and 919 m above sea level). The soil of the experimental area was classified as Dystrophic Red Latosol that has a clay texture. The chemical characterization of the soil follows: pH was 6.2; organic matter was 40 g dm^{-3}; P was 2.0 mg dm^{-3}; K was 75.0 mg dm^{-3}; Ca was 3.1 cmol$_c$dm^{-3}; Mg was 0.7 cmol$_c$dm^{-3}; H + AL was 2.6 cmol$_c$dm^{-3}; sum of bases was 4.1 cmol$_c$dm^{-3}; cation exchange capacity was 6.6 cmol$_c$dm^{-3}; and base saturation was 61%. The proportion of clay was 549 g kg^{-1}, and silt was 112 g kg^{-1}. The daily weather data for precipitation and temperature were obtained from a meteorological station that was located at 1.2 km from the experimental area (Figure 1), while daily total solar radiation data was obtained from the NASA POWER website (https://power.larc.nasa.gov/, accessed February 27, 2020).

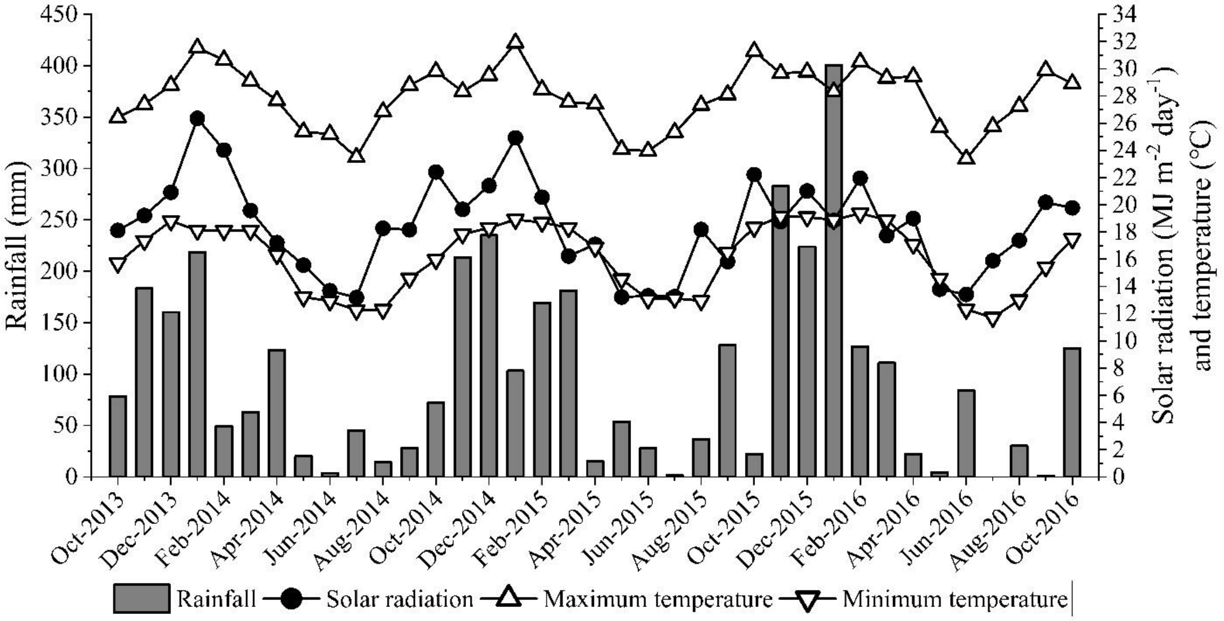

Figure 1. Average monthly total rainfall, solar radiation, and maximum and minimum temperature at the experimental site from 2013-2016 in Lavras, Minas Gerais, Brazil.

The experimental design was a randomized complete block with four replications. Treatments were five *Brachiaria* genotypes including three [*Brachiaria brizantha* (Hochst. Ex A. Rich.) Stapf.] (Marandu, Piatã and Xaraés), one *Brachiaria decumbens* (Stapf.) (Basilisk), and a three-way hybrid of *B. brizantha*, *B. ruziziensis* and *B. decumbens* (Mulato II). Grasses were sown at a rate of 6 kg ha^{-1} viable seed on April 2012 in 8-m by 4-m plots, and harvested at 15-cm stubble height every 28 d during the rainy season and every 42 d during the dry season. Agronomic responses were measured in two years, from October 2013 to November 2014 (Year 1) and from September 2015 to September 2016 (Year 3).

For Year 1, nitrogen and potassium were split-applied using $(NH_4)_2SO_4$ and KCl as fertilizers after each harvest, to provide a total annual application of 220 kg of N and 220 kg of K_2O ha^{-1}. From December 2014 to September 2015, plots were harvested and fertilized with the equivalent of 220 kg of N and 220 kg of K_2O ha^{-1} year^{-1}, but no data were collected. For Year 3, there were two levels of nitrogen fertilization, the non-fertilized treatment, equivalent to 0 kg of N ha^{-1} yr^{-1}, and the N-fertilized treatment, equivalent to 550 kg of N ha^{-1} yr^{-1} (both the treatments received the equivalent of 550 kg of K_2O ha^{-1} year^{-1}). See Cunha *et al.* (2022) for further details about this experiment.

2.2. Growth analysis measurements

At the end of each regrowth cycle, the forage inside two 0.5-m^2 quadrats per plot was clipped to a 15-cm stubble height and weighed fresh. From each herbage mass (HM) sample, defined as total biomass above stubble height, two subsamples of approximately 600 g were taken. One subsample was used to determine the dry matter (DM) concentration, while the other was used to determine the morphological composition of the plants, separating them into green leaf (leaf blade), stem (stem + leaf sheath) and dead material. The subsamples were dried in a forced ventilation oven at 55ºC for 72 h to calculate the DM. The annual herbage accumulation (HA) was obtained by summing HM across all regrowth cycles. After HM samples were taken, the entire plot was mechanically harvested at 15-cm stubble height and fertilized.

Stubble mass (SM) below 15-cm height was measured after HM harvest by sampling the biomass down to the soil level twice per year, on the longest day of the year (December 21) and on the shortest day of the year (June 21), to represent typical SM during the rainy and dry season regrowth cycles. From each SM sample, a sub-sample was taken to determine the DM concentration and the plant-part composition, separating them into green leaf, stem, and dead material.

Pre-harvest canopy height (CH) was measured on the last day of each regrowth cycle (immediately before plot harvest) by taking 24 readings using a light polyethylene sheet and a ruler.

2.3. Crop model: inputs and setup

The two-year dataset (Year 1 and Year 3) of the five genotypes grown under N-fertilization levels was prepared for simulations in DSSAT (Decision Support System for Agrotechnology Transfer) format. The process involved data entry for i) the daily meteorological data from the experimental site including solar radiation, maximum and minimum temperature, precipitation, dewpoint temperature, wind speed, and relative humidity; ii) the soil profile characterization data; iii) the crop management information; iv) the harvest timing information, the amount of stubble mass, the fraction of leaves present in the stubble mass (RSPLF) and the number of 're-staged' leaves (MVS) remaining in the stubble at harvest; and v) the measured experimental data, including the time series agronomic and plant-part composition, such as leaf, stem, herbage, and above ground shoot mass.

The description of the chemical, physical and hydrological characteristics of the soil profile related to soil water and nitrogen supply are important for the crop growth simulations. Also, detailed descriptions of the management conditions of the crop during the experiment were needed as inputs, such as planting method, prior crop residues, and the amount and timing of fertilizer application (Hoogenboom *et al.*, 2017).

For simulation of perennial crops, the CROPGRO-PFM model allows using a transplant option, which permits to initiate the simulation for an already established plant stand. In our study, the initial plant mass was set at 2,000 kg DM ha^{-1}, which was allocated to both above and belowground mass. In order to simulate soil organic carbon (SOC) in the soil horizons as well as the dynamics of a layer of surface residue above the topsoil, we used the CENTURY model (Parton *et al.*, 1988; Gijsman *et al.*, 2002) because it includes litter decomposition, dead root decomposition, and allows calibration of N mineralization by modifying the stable pool (SOM3) of the organic soil C module. Therefore, to initialize the CENTURY model, the ratio between SOM3 and total SOC for our simulations was 0.57, while the prior crop residue (residue of a prior perennial forage crop tilled into the soil) was 8,000 kg DM ha^{-1} with an N concentration of 10 g kg^{-1} (Pequeno *et al.*, 2017).

In our study the Penman-Monteith FAO 56 method (Allen *et al.*, 1998) was used to calculate potential evapotranspiration (ET) using wind speed and dewpoint temperature data of the weather file (Jones *et al.*, 2003). This method was chosen in our study, since, according to Saseendran *et al.* (2008), the Priestley and Taylor method tends to slightly overestimate ET. In addition, the Stage 1 soil water evaporation method of Ritchie (1972) was used for simulation of soil water evaporation.

The stubble mass was entered as the “MOW” value for the simulations and characterizes the unharvested living mass that remains after harvest clipping. The stubble mass and fraction of leaves were set based only on living tissue. In our work, the MVS parameter (hypothetical number of leaves left on the primary tiller axis after harvest used to “re-stage” partitioning) was kept at 3 (Lara *et al.*, 2012; Pequeno *et al.*, 2014). For the harvest dates when the stubble mass was not quantified, interpolation was used to estimate the mass between the dates when actual stubble mass was measured in the field.

2.4. Model improvement and evaluation

The CROPGRO-PFM model was calibrated and evaluated in five phases, always comparing the simulated data with the observed data:

- *Phase 1.* The default simulation was used to evaluate the ability of the CROPGRO-PFM model to simulate the field observations without any calibration using the input data of the experimental site, such as the weather data, the soil surface and profile characteristics, irrigation and fertilizer management, and initial conditions. For this phase, the model parameterization for the genetics coefficients, including species, ecotype, and cultivar, were those defined by Pequeno *et al.* (2017) for Marandu palisadegrass.

- *Phase 2.* The parameters related to the soil water supply, such as the lower limit and the drained upper limit, and rooting, such as the maximum rooting depth and the rooting shape, were calibrated. These parameters were adjusted to minimize water deficit, which was initially too severe during a prolonged period of drought as simulated during the default simulation.

- *Phase 3*. The parameters related to the N supply of the soil were calibrated. This included adjusting the parameters related to the mineralization of the soil organic matter and the residue, to improve the model simulations. The SOM3 parameter that defines the stable organic pool was adjusted for the non-fertilized and N-fertilized treatments.

- *Phase 4*. The genetic coefficients for ecotype and cultivar parameters were calibrated for each genotype. This phase was needed to evaluate our null hypothesis that the CROPGRO-PFM calibrated by Pequeno *et al.* (2017) for Marandu palisadegrass, can accurately simulate the growth and forage production of different *Brachiaria* genotypes under N-fertilization levels, with parameterization only of those specific characteristics related to the cultivar and ecotype parameters, and with no changes of the genetic coefficients that define the species traits. In this phase, the parameters related to the photosynthetic capacity of the genotype (LFMAX) and the intensity of allocation of assimilate to shoot versus storage tissues based on sensitivity to photoperiod (RDRMT) of the cultivar and ecotype file, were adjusted for each genotype.

- *Phase 5*. The genetic coefficients that define the species traits for *Brachiaria* were calibrated. This included the leaf-to-stem partitioning parameters for each genotype, along with modification of the LFMAX and RDRMT parameters for each genotype. This phase was instigated by a previous study (Cunha *et al.*, 2022) that found that there were significant differences ($P < 0.05$) in the fraction of leaf and stem among the genotypes. Therefore, it was assumed that each genotype should have its own species parameterization, along with distinct values for the cultivar and ecotype parameters. The measured values for shoot biomass, leaf and stem weight, herbage mass, and the fraction of leaves in shoot biomass were compared with the simulated values.

For the evaluation of the model performance after each one of the calibration phases, we compared the means of the simulated data with the measured data and the root-mean-square error (RMSE):

$$RMSE = \sqrt{\frac{1}{N}\sum_{i=1}^{N}(Y_i - \hat{Y}_i)^2}$$

where N is the total number of data points for comparison, Y_i is a given observed value, and $\hat{Y}_i$ is the corresponding value predicted by the model. A better model prediction will produce a smaller RMSE. In addition, we used the Willmott agreement index (D) (Willmott *et al.* 1985) which is given by:

$$D = 1 - \left[\frac{\sum_{i=1}^{N}\left(Y_i - \hat{Y}_i\right)^2}{\sum_{i=1}^{N}\left(\left|\hat{Y}_i - \bar{Y}\right| + \left|Y_i - \bar{Y}\right|\right)^2}\right], 0 \le D \le 1$$

where N is the number of observed data points, Y_i is a given observed value, $\hat{Y}_i$ is the corresponding value predicted by the model, and $\bar{Y}$ is the mean of the observed data. A D index near 1 indicates good model prediction.

3. Results

3.1. Simulation scenario

The two-year dataset (Year 1 and Year 3) of the five genotypes grown under N-fertilization levels were included in the model for the sequential simulation from 2013 to 2016. For Year 1, the measured data were the same for both non-fertilized and N-fertilized treatments, since the level of N that was applied was the same, i.e., 220 kg N ha^{-1} $year^{-1}$. For Year 3, the treatments received different levels of N-fertilization, equivalent to 0 and 550 kg N ha^{-1} $year^{-1}$. Therefore, the statistical results reported in tables and figures consider this scenario for the simulation of two treatments, i.e., the non-fertilized and the N-fertilized treatments.

3.2. Calibration of input parameters related to the water and N balance

In Phase 1 the default model (Pequeno *et al.*, 2017) was stimulated with the default inputs of soil water characteristics and soil organic carbon (Tables 1 and 2). In Phases 2 and 3, we adjusted the input parameters related to soil moisture and N mineralization, with adjustments made separately in Phase 2 to optimize soil moisture conditions and in Phase 3 to optimize N availability (Tables 1 and 2).

Table 1. Soil profile characteristics for soil water and N with both the initial and adjusted values used in Phases 1, 2, and 3.

Depth layer	Lower limit[1]	Drained upper limit[2]	Saturated upper limit[3]	Root growth factor[4]	Saturated hydraulic conductivity[5]	Initial soil water content[6]	Soil NH_4 content[7]	Soil NO_3 content[8]
(cm)	------------- *$cm^3\ cm^{-3}$*		-------------	*(0-1)*	*$(cm\ hr^{-1})$*	*$(mm^{3}\ mm^{-3})$*	--- *$g\ N\ Mg^{-1}$ soil*	---
Phase 1								
5	0.228	0.385	0.481	1.000	0.06			
15	0.228	0.385	0.481	1.000	0.06			
20						0.481	2.40	1.00
30	0.249	0.406	0.482	0.638	0.06			
40						0.481	3.50	1.90
45	0.249	0.406	0.465	0.472	0.06			
60	0.249	0.406	0.465	0.350	0.06			
70						0.482	1.70	1.40
90	0.308	0.456	0.468	0.223	0.06	0.465	0.90	1.10
110						0.465	1.00	1.00
120	0.207	0.341	0.452	0.122	0.06			
130						0.468	1.10	1.00
150	0.243	0.365	0.455	0.067	0.06			
160						0.452	0.90	1.00
180	0.259	0.361	0.457	0.037	0.06	0.455	0.90	0.70
200						0.457	0.80	0.80
210	0.259	0.361	0.457	0.020	0.06			
Phase 2 and 3								
5	0.244	0.385	0.481	1.000	0.09	0.385	2.40	1.00
15	0.244	0.385	0.481	1.000	0.09	0.385	2.40	1.00
20						0.406	2.40	1.00
30	0.265	0.406	0.482	0.800	0.09	0.406	3.50	1.90
40						0.406	3.50	1.90
45	0.265	0.406	0.465	0.600	0.09	0.406	1.70	1.40
60	0.265	0.406	0.465	0.400	0.09	0.406	1.70	1.40
70						0.406	1.70	1.40
90	0.265	0.406	0.468	0.300	0.09	0.406	0.90	1.10
110						0.406	1.00	1.00
120	0.265	0.406	0.452	0.200	0.09	0.406	1.10	1.00
130						0.406	1.10	1.00
150	0.265	0.406	0.455	0.180	0.09	0.406	0.90	1.00
160						0.406	0.90	1.00
180	0.265	0.406	0.457	0.105	0.09	0.406	0.90	0.70
200						0.406	0.80	0.80
210	0.265	0.406	0.457	0.070	0.09	0.406	0.80	0.80
240	0.265	0.406	0.457	0.050	0.09	0.406	0.80	0.80

Model parameter names: [1]SLLL, [2]SDUL, [3]SSAT, [4]SRGF, [5]SSKS, [6]SH2O, [7]SNH4, and [8]SNO3.

Table 2. Soil profile characteristics for soil organic C with both the initial and adjusted values used in Phases 1, 2, and 3.

Depth layer (cm)	Soil organic carbon (SOC, %)	*Phase 1*	*Phase 2*	*Phase 3*
		-------------- Soil stable organic carbon (SOM3, %) ---------------		
5	2.06	1.17	1.17	1.73
15	1.29	0.74	0.74	1.08
30	0.88	0.50	0.50	0.74
45	0.88	0.50	0.50	0.74
60	0.68	0.39	0.39	0.57
90	0.50	0.29	0.29	0.42
120	0.44	0.25	0.25	0.37
150	0.31	0.18	0.18	0.26
180	0.20	0.11	0.11	0.17
210	0.14	0.08	0.08	0.12
240	-	-	0.08	0.12

In Phase 2, the most significant modification made was to increase the depth of the soil profile, from 210 cm to 240 cm (Table 1). The available water in the individual soil layers was adjusted by the reducing the lower limit and modifying the drained upper limit parameter values (SLLL and SDUL, Table 1). The water runoff curve number of the soil (SLRO) was decreased from 73 to 61 to reduce the loss of water by runoff, since the presence of perennial forage as ground cover drastically reduces runoff potential and is responsible for increasing water infiltration into the soil. We increased the saturated hydraulic conductivity in the macropores (SSKS, from 0.06 to 0.09 cm h^{-1}), which also reduced runoff slightly (Table 1). We also increased the plant's potential rooting depth shape (SRGF) to mimic a deeply rooted perennial (Table 1). The parameters related to the initial soil water and N were kept the same as in Phase 1, however the lowest layer was assigned values to match the increased depth of the soil profile (Table 2).

In Phase 3, input parameters strictly related to the N balance in the soil and the decomposition of SOC (soil organic carbon) were adjusted. The SOM3 (soil stable organic carbon) set by Pequeno *et al.* (2017), resulted in a simulation with too much supply of mineralized N (SOM3/SOC ratio of 0.57). Therefore, we increased the value for SOM3 for each soil layer using the SOM3/SOC ratio of 0.84 (Table 2), with the goal to correctly mimic the reduced growth response of the non-fertilized compared to the N-fertilized treatments.

After the completion of Phases 2 and 3, we consider that the required input conditions related to water and N balance were satisfied. The effect of drought stress throughout the cool-dry season, mainly between June and October 2014, and between August and October 2015, was slightly reduced with the adjustments that were made (Figure 2, Phase 2). In addition, the N stress became more evident (red and light red lines) due to the adjustment to increase the stable fraction of organic carbon in the soil (Figure 2, Phase 3).

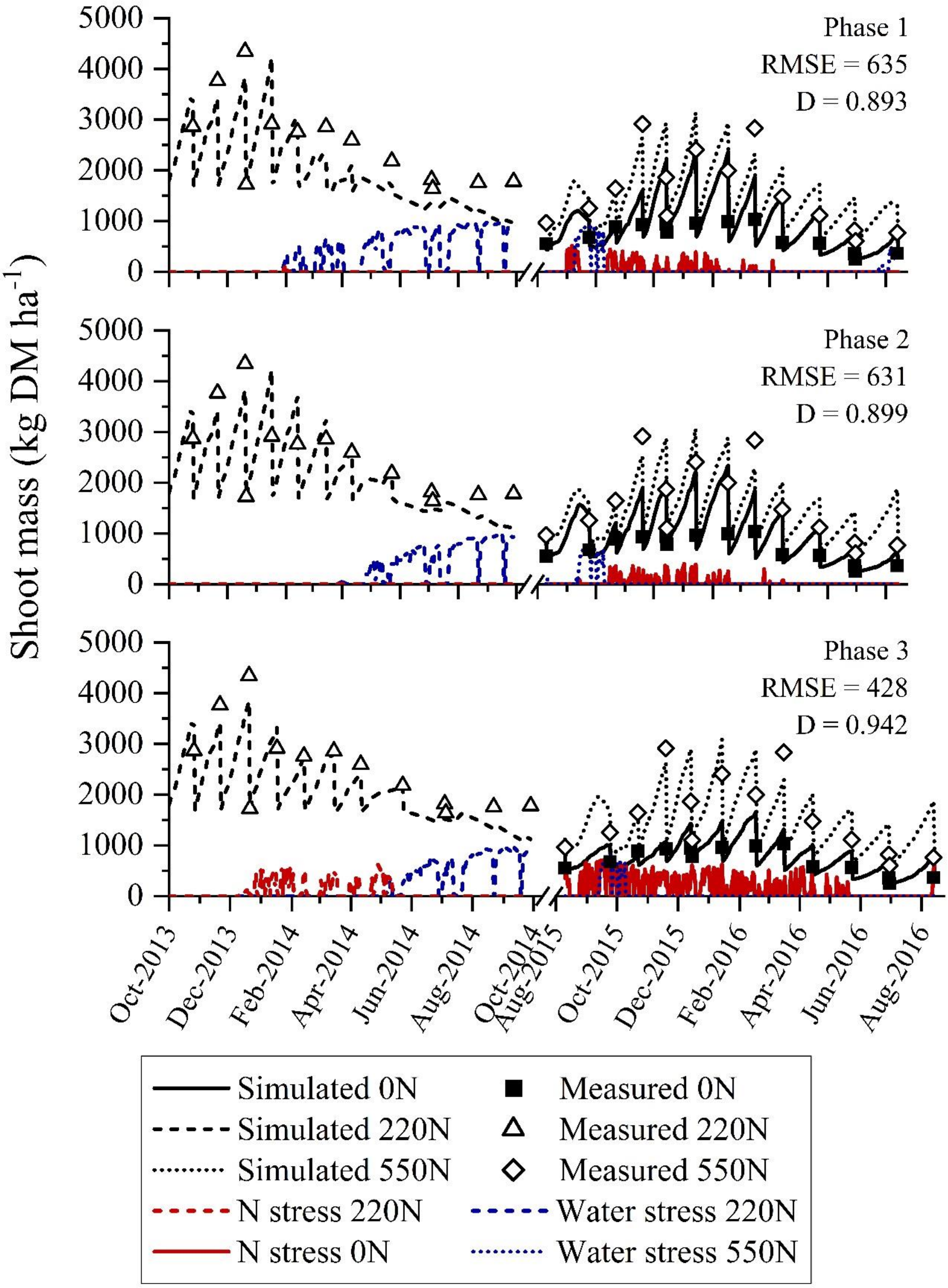

Figure 2. Comparison of measured (symbols) and simulated (lines) shoot biomass, water stress signal, and nitrogen stress signal over time, during Phases 1, 2, and 3 of calibration for Marandu palisadegrass growth under N-fertilization levels.

Overall, assuming that all *Brachiaria* genotypes respond similarly, the adjustments ensured a good simulation of the regrowth cycles and maximized the model's performance. The main characteristics such as shoot biomass, herbage, leaf fraction, leaf mass, and stem mass, were simulated with greater precision, and approximated the simulated average values to those measured, reducing the RMSE values and increasing the Wilmot agreement index values (Table 3).

Table 3. Means and statistics for the growth analysis for all genotypes (five) and N-fertilization levels (two) combined (n=10) in Phases 1, 2, and 3.

Variables	Meas.	*Phase 1*			*Phase 2*			*Phase 3*		
		Sim.	RMSE	D	Sim.	RMSE	D	Sim.	RMSE	D
Shoot, kg DM ha^{-1}	1,833	1,940	616	0.898	2,081	607	0.905	1,907	414	0.945
Herbage, kg DM ha^{-1}	885	1,181	690	0.744	1,349	710	0.729	1,098	456	0.850
Leaf in shoot, %	60.0	62.7	8.3	0.797	63.3	8.5	0.790	61.3	7.5	0.810
Leaf, kg DM ha^{-1}	1,233	1,366	527	0.838	1,478	521	0.842	1,294	379	0.897
Stem, kg DM ha^{-1}	715	719	214	0.897	768	217	0.900	746	180	0.922

Measured (Meas.), Simulated (Sim.), Root mean square error (RMSE) and Wilmot agreement index (D), are averaged over N-fertilization treatments.

In Phase 3, the average shoot biomass for all five genotypes and treatments (non-fertilized and the N-fertilized) was slightly overestimated (by 4%). The herbage was still overestimated when compared with the measured values (24%). The fraction of leaves present in the shoot biomass was also slightly overestimated. Shoot mass, leaf mass and stem mass showed considerable improvement with reduction in RMSE and improved D-statistic when the simulated values were compared with the measured values (Table 3).

3.3. Calibration of parameters related to the plant growth dynamics

In Phase 4, we obtained the best values for the LFMAX and RDRMT parameters for each genotype. As the model after Phase 3 somewhat overestimated some of the biomass values, the best combination of values for both parameters was achieved by slightly reducing the LFMAX parameter values (from 1.80 to as low as 1.60) and increasing the RDRMT parameter values (from 0.475 to as high as 0.555) (Table 4, Phase 4). This decreased the yield of genotypes and at the same time optimized the redirection of photo-assimilates to plant storage tissues. These adjustments were made considering the variations in shoot biomass and herbage of each genotype.

Table 4. Default cultivar and ecotype parameters values used in Phases 1, 2 and 3, and adjusted parameter values obtained in Phases 4 and 5.

Parameter	Definition	Genotype	Parameter values[*]		
			Phase 1, 2 and 3	*Phase 4*	*Phase 5*
LFMAX	Maximum leaf photosynthesis rate at 30 °C, 350 vpm CO_2, and high light (mg CO_2 m^{-2} s^{-1}).	*Basilisk*	**1.80**	1.60	1.60
		Marandu		1.60	1.60
		Mulato II		1.70	1.68
		Piatã		1.60	1.60
		Xaraés		1.80	1.80
RDRMT	Relative dormancy sensitivity of the genotype to daylength to partitioning.	*Basilisk*	**0.475**	0.515	0.515
		Marandu		0.490	0.540
		Mulato II		0.515	0.515
		Piatã		0.480	0.495
		Xaraés		0.555	0.505

[*] Values in bold correspond to the default values of the cultivar and ecotype parameters (Pequeno *et al.*, 2017).

The effort in Phase 4, with the objective of optimizing the values of the LFMAX and RDRMT parameters for each *Brachiaria* genotype, improved the statistical performance of the simulations, bringing the simulated values closer to those measured (lower RMSE and upper D). Considering the mean of genotypes and treatments (Table 5, Phase 4), shoot biomass was slightly overestimated (0.5%). Also, herbage was still overestimated when compared to measured values (by 14.6%). The fraction of leaves present in the shoot biomass was slightly overestimated by the model. The statistics for simulated leaf mass and stem mass were improved and simulated values were very close to the measured values.

Table 5. Means and statistics for the growth analysis for all genotypes (five) and N-fertilization levels (two) combined (n=10) in Phase 4 and 5.

Variable	Meas.	*Phase 4*			*Phase 5*		
		Sim.	RMSE	D	Sim.	RMSE	D
Shoot, kg DM ha^{-1}	1,833	1,842	407	0.947	1,836	410	0.947
Herbage, kg DM ha^{-1}	885	1,015	431	0.859	1,010	434	0.857
Leaf in shoot, %	60.0	60.8	7.5	0.812	60.5	7.0	0.831
Leaf, kg DM ha^{-1}	1,233	1,234	372	0.901	1,228	367	0.903
Stem, kg DM ha^{-1}	715	730	175	0.927	730	161	0.941

Measured (Meas.), Simulated (Sim.), Root mean square error (RMSE) and Wilmot agreement index (D), are averaged over N-fertilization treatments.

In Phase 5 we modified the partitioning function for each genotype, aiming to improve the estimates of the leaf as a fraction of the shoot biomass, as well as the total amount of leaf and stem mass (Table 6). In this phase, the values of the LFMAX and RDRMT parameters were also recalibrated (Table 4, Phase 5). The partition coefficients between leaves and stem, defined in the crop species file, were adjusted for each *Brachiaria* genotype (Table 6) to improve the statistical performance of the simulation of leaf fraction in shoot biomass, leaf, and stem mass (Table 5, Phase 5). The sum of the values of the partition coefficients for YLEAF and YSTEM remained the same, so that the increase in assimilates allocated to leaves was at the expense of assimilates allocated to stems.

Table 6. Default species parameter values (in bold) used in Phases 1, 2, 3 and 4, and adjusted values obtained in Phase 5.

Parameter	Definition	Genotype	Parameter values[*]							
XLEAF	Cumulative leaf number on main axis at which the partitioning is defined (linked to YLEAF, YSTEM, YSTOR and YROOT).		**0.0**	**2.0**	**3.0**	**5.0**	**7.0**	**10.0**	**30.0**	**40.0**
YLEAF	Daily partitioning of assimilate to leaves as a function of the vegetative stage.		**0.80**	**0.80**	**0.72**	**0.63**	**0.52**	**0.51**	**0.50**	**0.50**
		Basilisk	0.76	0.76	0.68	0.59	0.48	0.47	0.46	0.46
		Marandu	0.79	0.79	0.71	0.62	0.51	0.50	0.49	0.49
		Mulato II	0.82	0.82	0.74	0.65	0.54	0.53	0.52	0.52
		Piatã	0.78	0.78	0.70	0.61	0.50	0.49	0.48	0.48
		Xaraés	0.84	0.84	0.76	0.67	0.56	0.55	0.54	0.54
YSTEM	Daily partitioning of assimilate to stems as a function of the vegetative stage.		**0.10**	**0.10**	**0.14**	**0.17**	**0.32**	**0.36**	**0.35**	**0.35**
		Basilisk	0.14	0.14	0.18	0.21	0.36	0.40	0.39	0.39
		Marandu	0.11	0.11	0.15	0.18	0.33	0.37	0.36	0.36
		Mulato II	0.08	0.08	0.12	0.15	0.30	0.34	0.33	0.33
		Piatã	0.12	0.12	0.16	0.19	0.34	0.38	0.37	0.37
		Xaraés	0.06	0.06	0.10	0.13	0.28	0.32	0.31	0.31
YSTOR	Daily partitioning of assimilate to storage tissues as a function of the vegetative stage.		**0.01**	**0.01**	**0.03**	**0.04**	**0.04**	**0.04**	**0.04**	**0.04**
YROOT	Daily partitioning of assimilate to roots as a function of the vegetative stage.		**0.09**	**0.09**	**0.11**	**0.16**	**0.12**	**0.09**	**0.11**	**0.11**

[*] Values in bold correspond to the default values of the species parameters (Pequeno *et al.*, 2017).

There was relatively small adjustment in the partitioning coefficients for YLEAF and YSTEM (Table 6). For Basilisk, Marandu and Piatã, the leaf partitioning values were reduced by as much as 0.04, indicating that more of the photo-assimilates are directed to the stems of those genotypes. The partition of assimilates to other tissues, such as storage tissues and roots, remained unchanged.

Figure 3 provides a graphical representation of the adjustment made to partitioning functions to leaf, stem, root, and storage for each genotype compared to the standard defined by Pequeno *et al.* (2017) as a function of advancing vegetative stage (equivalent to crop regrowth age). This figure also illustrates that the simulated fraction partitioned to the storage tissues and the roots was not changed across genotypes, admittedly because of lack of data in this trial for parameterization.

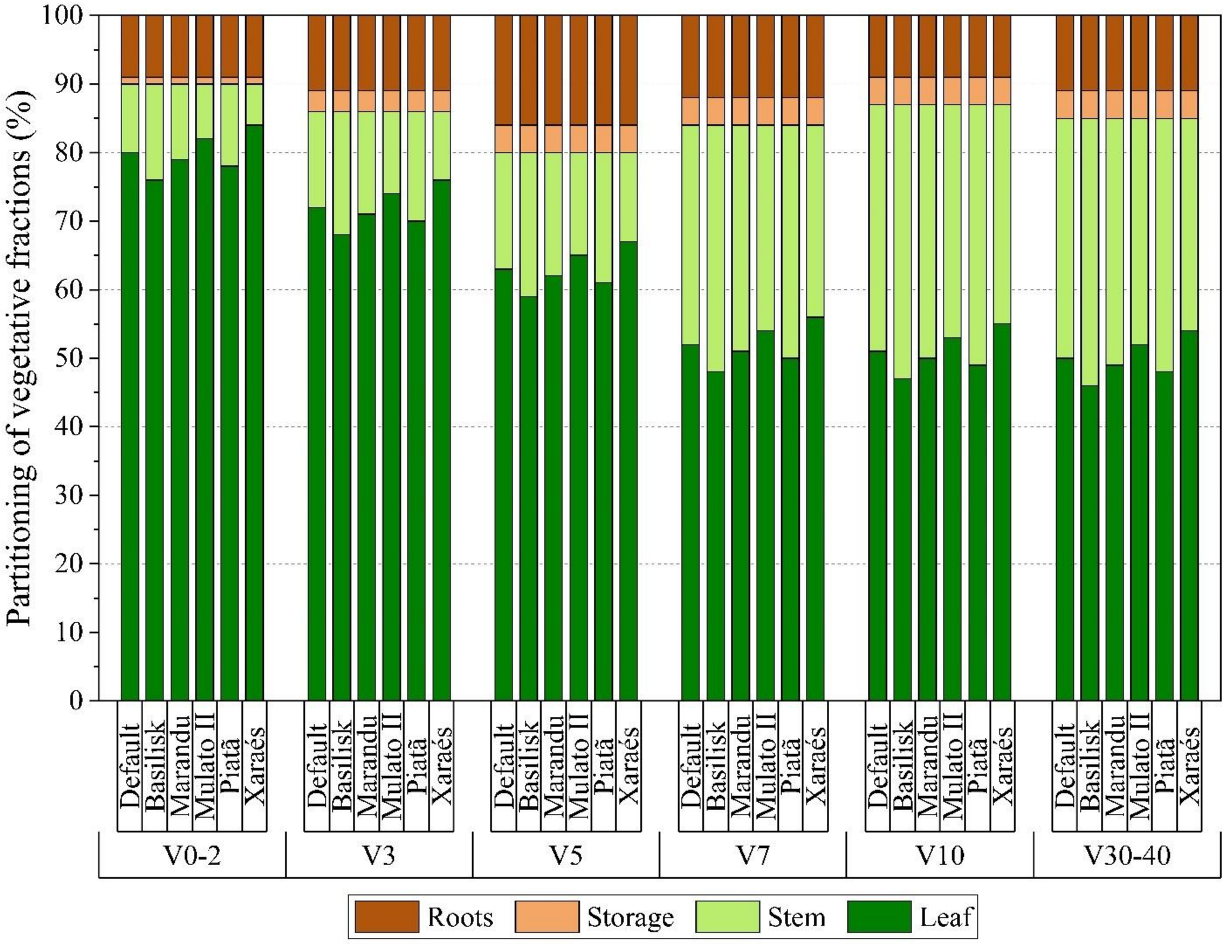

Figure 3. Default and adjusted partitioning parameter values for successive V-stages for each *Brachiaria* genotype obtained in Phase 5.

Regarding any further adjustments to the genetic coefficients in Phase 5, only Marandu required a slight reduction (from 1.70 to 1.68 mg CO_2 m^{-2} s^{-1}) in the value of LFMAX, indicating that the maximum photosynthesis rate of this genotype had to be reduced slightly to reflect the differences between the simulated and the measured yield of biomass, leaf, and stem. The RDRMT parameter was increased by 0.50 for Marandu and by 0.15 for Piatã, indicating that the allocation of photo-assimilates to reserve tissues should be greater for these species, while for Xaraés this value was reduced by 0.50 (Table 4, Phase 5).

3.4. Model improvement allows contrasting different genotypes

One of the objectives of Phase 5 was to evaluate the ability of the CROPGRO-PFM model to simulate growth differences and individual morphological aspects of each *Brachiaria* genotype. The combination of the partitioning function adjustments (Table 6), associated with the adjustment of the LFMAX and RDRMT parameters for each genotype, improved the ability of the CROPGRO-PFM to simulate the differences among the genotypes. With this improvement, the model simulated the leaf and stem mass, herbage yield and the fraction of leaves in shoot with satisfactory performance (D > 0.831).

Analyzing the statistical results and the adjustment of the simulated growth variables for each genotype, calculated over the non-fertilized and N-fertilized treatments, we can document the importance of the fine adjustments made up to Phase 5, where all the genotypes had improved statistical fit (low RMSE and high D) (Table 7).

Table 7. Means and statistics of time-series performance for simulated growth variables after separate species parameter calibration for each *Brachiaria* genotype (Phase 5).

Variables	Genotype	Measured	Simulated	RMSE	D
Shoot, kg DM ha^{-1}	*Basilisk*	1,592	1,586	370	0.955
	Marandu	1,811	1,816	409	0.947
	Mulato II	1,933	1,941	409	0.943
	Piatã	1,937	1,971	449	0.948
	Xaraés	1,891	1,866	413	0.941
Herbage, kg DM ha^{-1}	*Basilisk*	881	955	393	0.888
	Marandu	843	976	428	0.858
	Mulato II	859	1,019	418	0.866
	Piatã	873	1,038	494	0.822
	Xaraés	970	1,062	439	0.852
Leaf in shoot, %	*Basilisk*	57.2	57.4	5.9	0.850
	Marandu	59.1	59.5	7.3	0.829
	Mulato II	63.4	64.4	7.2	0.797
	Piatã	55.6	56.6	7.4	0.808
	Xaraés	64.4	64.8	7.5	0.871
Leaf, kg DM ha^{-1}	*Basilisk*	1,010	1,002	270	0.929
	Marandu	1,182	1,192	365	0.899
	Mulato II	1,373	1,376	384	0.904
	Piatã	1,223	1,230	399	0.898
	Xaraés	1,378	1,339	417	0.886
Stem, kg DM ha^{-1}	*Basilisk*	701	701	212	0.926
	Marandu	738	742	205	0.916
	Mulato II	671	689	132	0.932
	Piatã	826	865	138	0.969
	Xaraés	641	655	117	0.961

Root mean square error (RMSE) and Wilmot agreement index (D), are averaged over N-fertilization treatments.

In general, the shoot biomass was well simulated after calibration, as demonstrated by the small differences between the simulated and measured values for each genotype. The simulated average over all five genotypes was 1,833 kg DM ha^{-1} compared to an average measured of 1,836 kg DM ha^{-1}. The RMSE was 410 kg DM ha^{-1}, which indicates a good accuracy, and is also supported by a high average D value of 0.947. The simulated herbage mass was overestimated when compared with the measured average (1,010 vs. 885 kg DM ha^{-1}) with a RMSE was 434 kg DM ha^{-1}, while the value for D was 0.857.

Simulating periodic herbage production over time is very important, especially for grazing applications of the model. The simulated herbage showed good agreement with measured herbage, across low to high N-fertilization levels (Figure 4). These 1:1 graphs indicate that the agreement between the simulated and measured values is good, although there was greater variability of the measured data among the N-fertilized cases, a common result to other studies that evaluate the effect of different levels of N-fertilization on the herbage production, and canopy structure and architecture (Yasuoka *et al.*, 2017; Delevatti *et al.*, 2019).

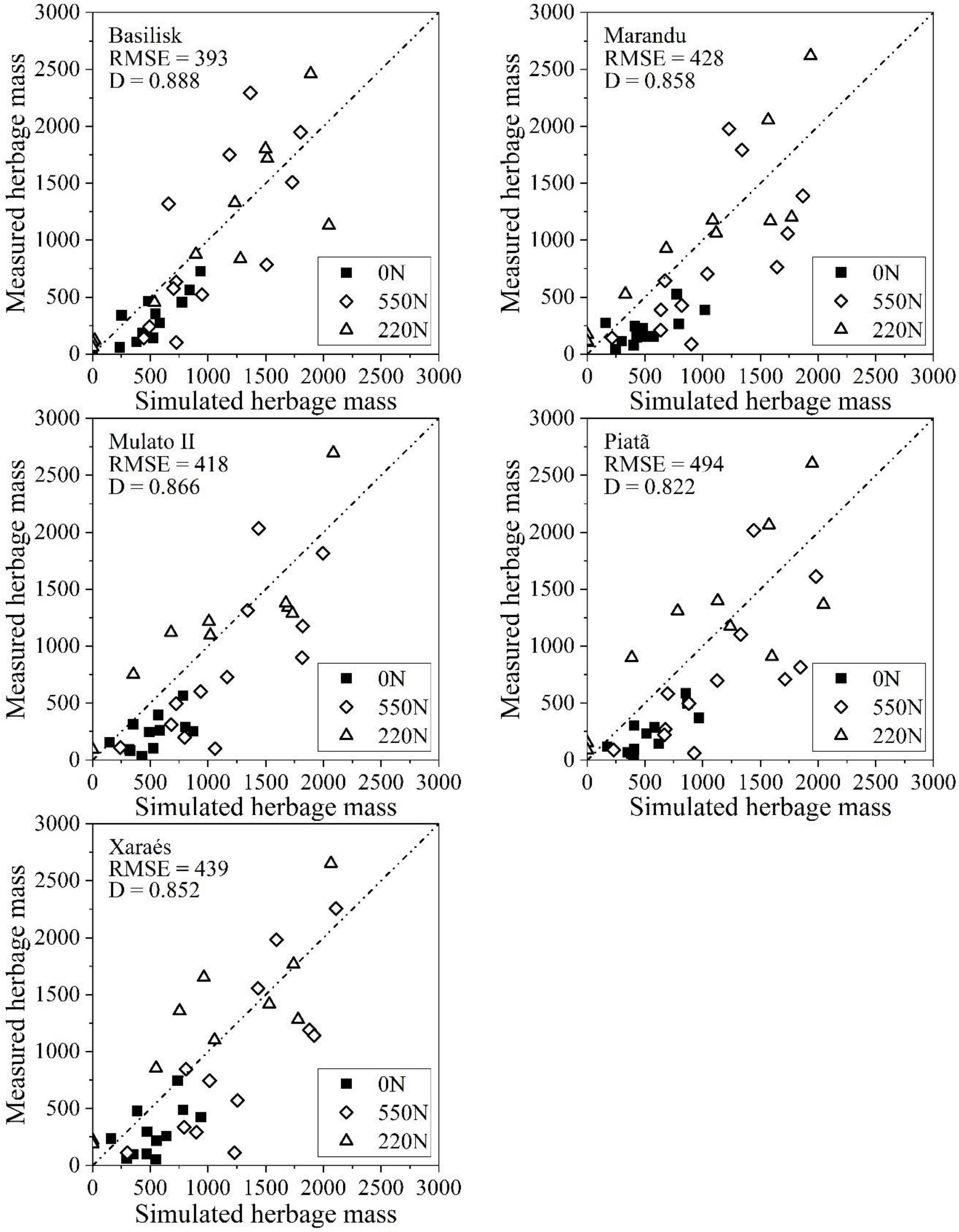

Figure 4. Relationship between measured and simulated herbage mass (kg DM ha^{-1}) after Phase 5 for each *Brachiaria* genotype, under N-fertilization levels.

Following model improvement and calibration, the fraction of leaves in the shoot over time was well simulated, as demonstrated by the simulated and measured values for each genotype (Figure 5). The fraction of leaves in the shoot at each harvest, had a difference of just 0.5% between the simulated and the measured averages, indicating good adjustment and accuracy (Table 5). Mulato II and Xaraés had a higher fraction of leaves in the shoot than Basilisk, Marandu and Piatã (63.9% vs. 57.3%), indicating their greater production potential.

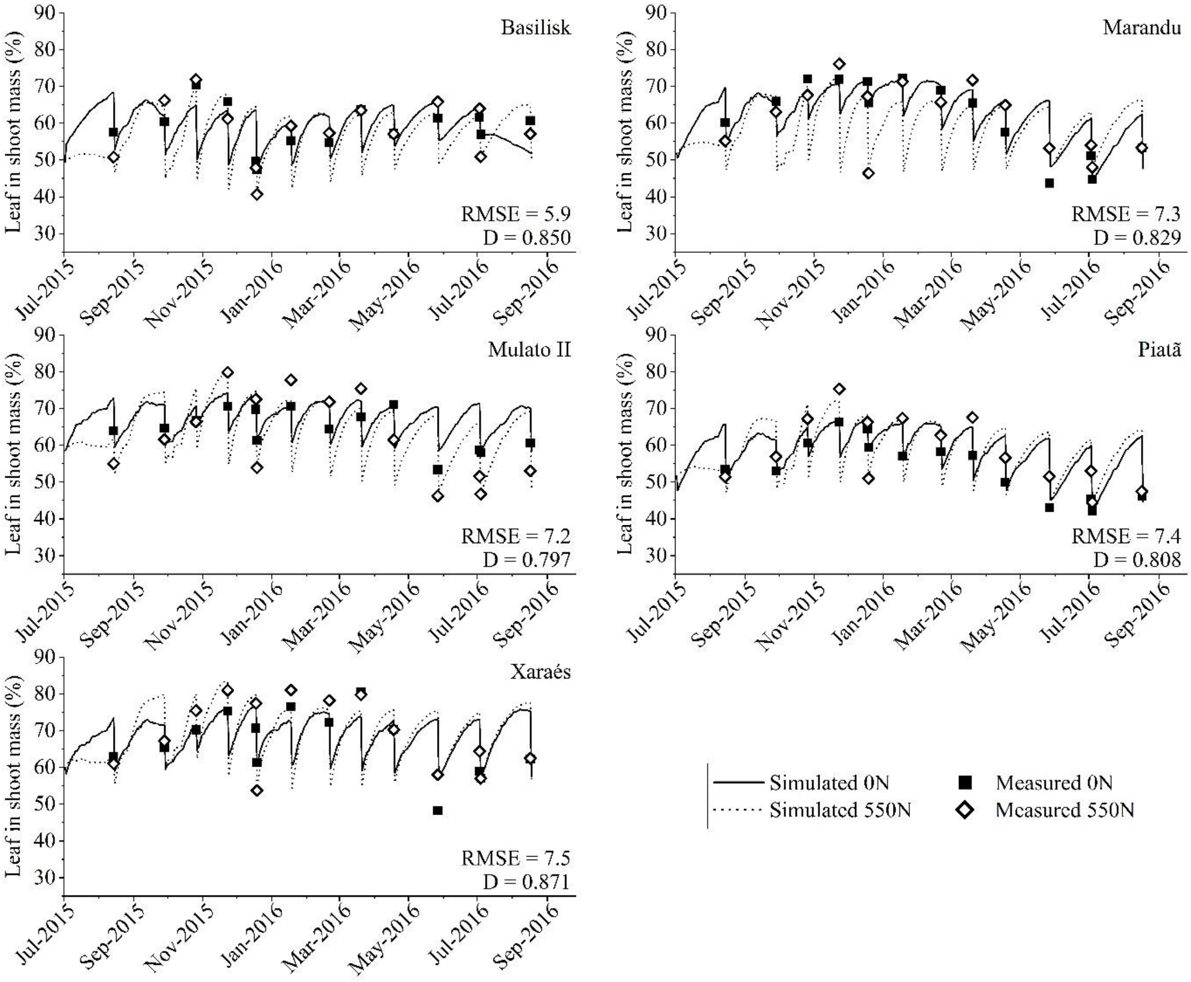

Figure 5. Measured (symbols) and simulated (lines) leaf fraction of shoot mass for each *Brachiaria* genotype, under N-fertilization levels for Year 3 (July, 2015 – September, 2016).

The leaf mass was also well simulated, as demonstrated by the simulated and measured values for each genotype (Figure 6). The leaf mass at each harvest, had a difference of just 7 kg DM ha^{-1} between the simulated and the measured averages, indicating good adjustment and accuracy (Table 5). The harvest cycles during the rainy season had the largest production of herbage and leaves, sufficient to differentiate the genotypes. Mulato II and Xaraés had a higher leaf production than Basilisk, Marandu and Piatã (1,375 kg DM ha^{-1} vs. 1,138 kg DM ha^{-1}), indicating an increase of almost 21% when comparing the most and least productive group.

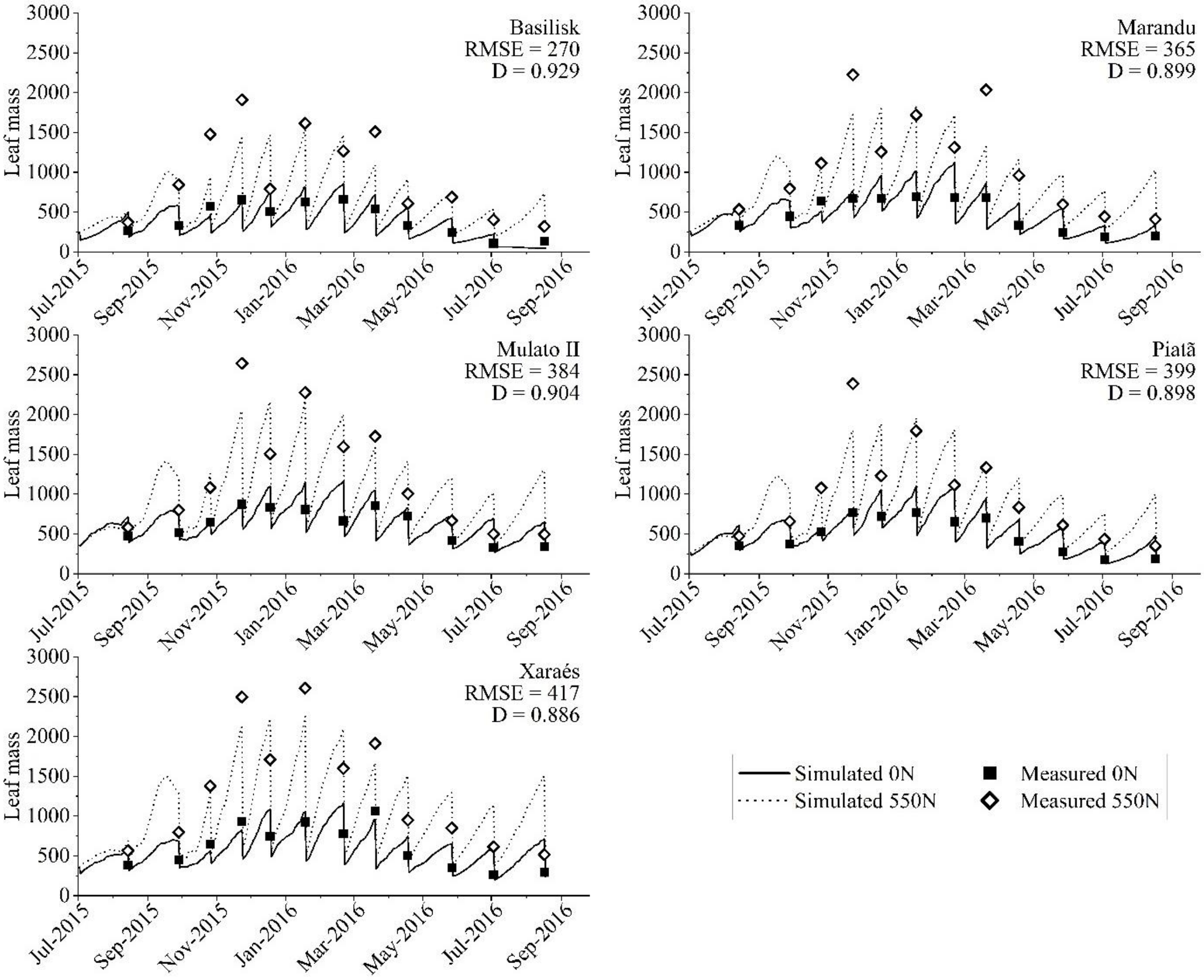

Figure 6. Measured (symbols) and simulated (lines) leaf mass (kg DM ha^{-1}) for each *Brachiaria* genotype, under N-fertilization levels for Year 3 (July, 2015 – September, 2016).

Similar to the leaf component, the stem mass at each harvest was also well simulated (Table 5). The stem mass had a simulated average value of just 15 kg DM ha^{-1} more than the measured average (Figure 7). Stem growth also was greater during the rainy period of the year.

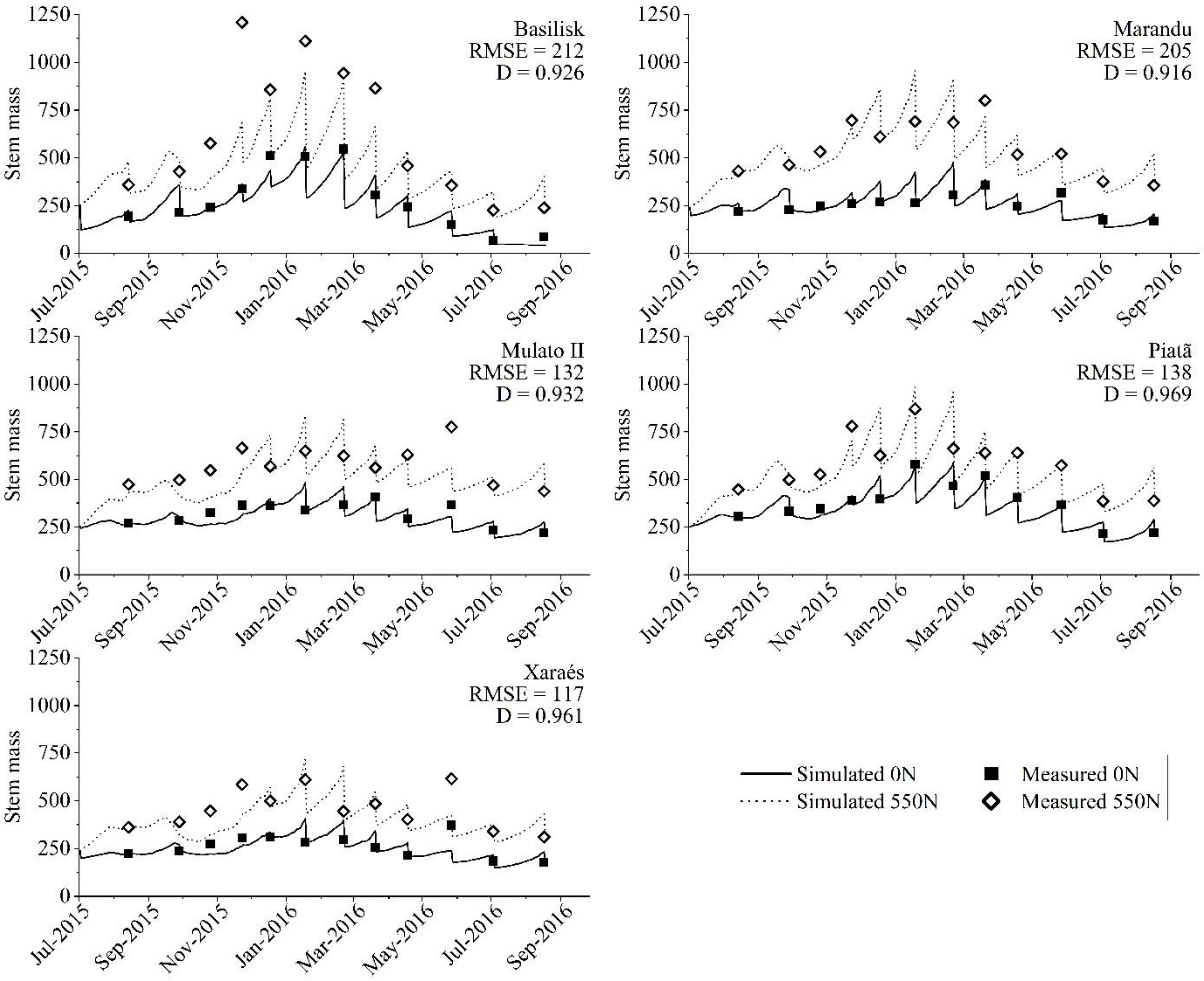

Figure 7. Measured (symbols) and simulated (lines) stem mass (kg DM ha^{-1}) for each *Brachiaria* genotype, under N-fertilization levels for Year 3 (July, 2015 – September, 2016).

4. Discussion

After each calibration phase, the overall performance of the CROPGRO-PFM model estimates was evaluated (Tables 3 and 5). Initially, the simulated growth was somewhat higher than the measured values (Table 3, Phase 1). The adjustments to the input parameters made in Phase 2 reduced the effects of water stress, mainly characterized by an excessive reduction in the average growth of genotypes compared to the average growth observed during the cool-dry season. Despite the improvement in the statistical performance of the model (Table 3, Phase 2), these adjustments promoted an excessive increase in plant growth and, consequently, in the average biomass production for the years that were used for evaluation and not just in the cool-dry season. Therefore, in Phase 3 we adjusted the total biomass production across the low versus high N fertilization treatments by modifying the soil N availability parameters. We increased SOM3 content in the soil (holding total SOC constant) which reduced the rate of N mineralization, thus resulting in a slight decrease in plant productivity. These combined actions improved the statistical performance, and the simulated values were close to the measured values of the variables related to productivity and the morphological composition of the plants (Table 3, Phase 3).

The adjustments made in Phases 2 and 3 were related to the water and N balance. This reduced the effect of water stress in Phase 2 and increased the N stress signal in Phase 3 over time, mostly to reduce the productivity of the zero N treatment (Figure 2). This outcome is a result of the reduction of the N mineralization rate to give the correct simulation of the responses of non-fertilized and the N-fertilized treatments, especially reducing the growth of the non-fertilized treatment.

After the required input conditions had been satisfied, the following calibration phases were used to evaluate the hypothesis of the model's ability to simulate the growth of each *Brachiaria* genotype, adjusting the genetic parameters for each genotype to improve the model performance. In contrast to the many phenology and seed growth parameters of typical annual seed-producing models, only the LFMAX parameter of the cultivar file and RDRMT parameter of the ecotype file proved to be relevant for the CROPGRO-PFM model. The LFMAX describes the leaf photosynthetic capacity, and the RDRMT parameter describes the intensity of allocation of assimilate to shoot versus storage tissues based on sensitivity to photoperiod. Therefore, in Phase 4, there was a small reduction of LFMAX combined with an increase of the RDRMT parameter (Table 4), indicating that the rate of photosynthesis was not drastically reduced, remaining within the physiological range of forage grasses (Lara and Pedreira, 2011).

Despite the improvement in model performance (Table 5, Phase 4), the adjustments made were still not sufficient to correctly mimic the daily partitioning of photo-assimilates between leaf and stem of individual *Brachiaria* genotypes, as reported by Cunha *et al.* (2022). The growth analysis performed by Cunha *et al.* (2022) suggested the need to model differences in the partitioning and plant-part composition of genotypes during regrowth. Based on this knowledge, we focused on modifying key parameters that affect the fraction of leaves in the shoot, as well as the production of leaf and stem mass over time. Thus, in Phase 5 we modified the partitioning function (Table 6) for each genotype (each genotype having its own species file) and recalibrated the values of the LFMAX and RDRMT parameters (Table 4, Phase 5). The goal was to improve the estimates of the leaf as a fraction of the shoot biomass, as well as the total amount of leaf and stem mass, assuming that each genotype should have its own set of genetic (species) coefficients, along with distinct cultivar and ecotype parameter values.

The CROPGRO-PFM model partitioning functions for allocation to leaves (YLEAF), stems (YSTEM), storage tissues (YSTOR) and roots (YROOT) vary with the vegetative growth stage (XLEAF, main axis node number per tiller) and are additionally modified by water deficit and N deficiency. For the simulation of these perennial grasses, changes in partitioning dynamics occur mainly between vegetative stages 3 and 7 (Figure 3), when main tillers have three to seven live leaves in which the progression of the V-stage is mainly modulated by thermal time. This range of variation in the number of live leaves is one of the tools used to manage the growth and harvest of grasses as reported in the literature (Donaghy and Fulkerson, 2001).

Compared to the pattern previously established for Marandu by Pequeno *et al.* (2017), there are small differences in the partition function values for each of the five genotypes (Figure 3). However, these were required to express the differences among genotypes of the same genus. Mulato II and Xaraés required an increase in leaf partition function values, indicating that these genotypes tend to direct more of the photo-assimilates to the leaves at the expense of the stem (Cunha *et al.*, 2022). Furthermore, Cunha *et al.* (2022) reported that the main contrast occurred between Xaraés and Basilisk, over the average of N-fertilized and non-fertilized genotypes, Xaraés had a leaf:stem ratio that was 4.3 times greater than Basilisk (leaf:stem of 12.8 vs. 2.4). This confirms the difference in assimilate partitioning among leaf and stem for the evaluated genotypes.

The variation in phenology and flowering time of perennial forage grasses also impacts the morphological plant-part composition. The appearance of inflorescences at specific times of the year is accompanied by an increase in height and hence the total amount of stem mass that is produced compared to other aboveground plant components (Cunha *et al.*, 2022). These factors, mainly modulated by photoperiod, promote the reduction of leaf growth, and could influence the quality and nutritional value of the forages. With the model improved for simulating the daily partitioning of assimilate to leaves, we can now assess whether the morphological and structural variations of individual genotypes can be a key factor in recommending their use, as well as helping farmers identify the most promising genotypes depending on the farm conditions and forage-livestock-system.

The CROPGRO-PFM model successfully simulates shoot biomass production, leaf as a fraction of the shoot, and the leaf and stem production throughout the year (Table 7, Figures 6, 7 and 8), thus reproducing the results of the growth analysis reported by Cunha *et al.* (2022). The intensity of the harvest frequency to 15-cm stubble height can influence growth, as well as the structural characteristics of the canopy, thus resulting in different plant-part composition of the genotypes (Pedreira *et al.*, 2017). At the same time, it demonstrates that the model well simulates the leaf fraction in the shoot, as an important indicator of forage quality (De Pinho Costa *et al.*, 2014; Pedreira *et al.*, 2017), because leaf fraction is associated with higher productivity, nutritional value, and potential use as a forage resource (Vendramini *et al.*, 2014). Furthermore, the leaf fraction in the shoot was a critical factor for the parameterization of the model, as it helps to adjust the partition of photo-assimilates that are directed to the vegetative portion of each genotype.

Simulating the dynamics of the leaves and stems is extremely important for the application of simulation models for grazing management, and especially with respect to production planning and use of forage resources on farms. With the adjustments of different values of species, cultivar and ecotype parameters for each genotype, the model was able to satisfactorily simulate the growth of the different *Brachiaria* genotypes in terms of herbage and the partitioning between leaves and stems (Table 5, Phase 5). This approach increases the value of using the CROPGRO-PFM model to predict the growth of different genotypes of the genus *Brachiaria* (Table 7).

At present, the partitioning function is in the species file, but we believe it will be possible to add code and a new single parameter to the cultivar file that modifies the function in the species file responsible for distribution of photo-assimilates between leaves and stems for different genotypes of *Brachiaria*. It is important to note that such a parameter can also be used to express small differences in the partitioning for genotypes among other forage grass species, without the need for a new species file for each new genotype. Adding this improved genetic functionality only requires a “step-type” modification of the partitioning function between the leaf and stem fractions. A modification of the code is anticipated in the near future to move this partitioning control function into either the ecotype or cultivar file for the convenience of model users.

5. Conclusions

The CROPGRO-PFM model was evaluated and improved by: i) the good parameterization of the model input data related to the water and N balance in the soil, focusing on properly defining the inputs, such as the soil water supply characteristics (SLLL, SDUL, rooting) and the stable soil carbon pool (SOM3) for N mineralization, which are important for an accurate

N response; and ii) the adjustment of the values of the parameters related to the photosynthetic capacity, partition between leaf and stem organs, and the intensity of the allocation of photo-assimilates to reserve tissues based on sensitivity to the photoperiod.

With the improvement of the current model, it will be possible to simulate seasonal growth and determine the morphological composition of the "plant-part" over time. This information will be useful in forage planning and will assist in the selection of forage genotypes for different production systems, based on the specificities of each grass. This study, therefore, makes the CROPGRO-PFM model even more suitable as a decision support tool for tropical and sub-tropical regions helping farmers to evaluate the efficiency of their own forage-livestock-systems.

Author Contributions

Conceptualization, methodology and supervision, M.L., K.B. and G.H.; investigation and data curation, D.C. and L.S.; formal analysis and writing – original draft, D.C. and S.S.; writing – review and editing, all authors.

Declaration of competing interest

The authors declare that there is no conflict of interest.

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SECOND SECTION

ARTICLE 3

WhenHarvest, a web-based forecasting system to determine the expected harvest time under different climate scenarios: a case study with Marandu palisade grass

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Abstract

Different simple and robust predictive models were built to simulate the growth and development of plants over the years. However, the use of such models by farmers is still limited, due to the need to insert several variables to configure the simulation scenario. Therefore, a model widely discussed in the literature adopted to represent the non-chronological development time of crops based on the thermal sum of growing degree-days (GDD) was incorporated into a web tool. The WhenHarvest forecasting system was formulated to predict the optimal time to conduct the first harvest of Marandu palisade grass after sowing (grown in the absence of water and nutritional deficit) and it can be adapted to predict the harvest for different crops. The farmer only needs to inform the location of his/her crop, the sowing date and some soil attributes, and the system will return the probable date of the harvest considering different scenarios of sowing date and climate. Users will also receive complementary information related to the water balance of the period under study. The system can be used to predict the harvest of those who have already sowed or to plan the best time for sowing.

Keywords: Agrometeorological models; Growth simulation; Weather conditions; Plant phenology.

1. Introduction

Plant growth requires light, water, carbon dioxide, mineral nutrients, and adequate temperature. These demands involve the environment and plant physiology and are related to i) meteorological factors such as light, photoperiod, temperature, precipitation, humidity, and wind; ii) edaphic factors such as site topography, slope, exposure, and soil properties; and iii) biotic factors such as pests, and diseases [1]. Given this complexity, several initiatives were launched to develop mathematical models with the objective of expressing the growth and development of plants, associating factors related to the environment and physiology. There are numerous models described in the literature, with different degrees of complexity and type of temperature response function [2]. Widely spread and accepted in the scientific community, the growth and development of plants is related to the ambient temperature. One of the most common ways of expressing this relationship is based on the sum of growing degree-days (GDD) [3].

The concept of GDD is a widely used tool for agricultural practices [4, 5]. It also allows real-time monitoring of the phenological development of the crop, using a thermal time scale, a more reliable representation than chronological time [6]. Despite being a low-complexity model, it is still little used in practice to support farmers decision-making. Mainly due to the need to perform the calculations, considering the local temperature data and the basal temperature of the plant development [7]. In addition, there are no simple tools that estimate future values for the accumulation of GDD and at the same time bring with them the ability to simulate scenarios that include climatic variations and information related to water availability in plant-atmosphere system. Therefore, we developed an alternative way that can help farmers make real-time decisions.

In this article, we present an alternative web-based forecasting system to calculate and predict GDD accumulation and water balance that can help farmers plan the agricultural calendar, called WhenHarvest. The system was designed to determine the optimal harvest time, and recommendations for the best planting season and other activities related to soil analysis and fertilization. The developed solution applies a serverless approach, exploiting a set of cloud-based microservices. This type of approach allows for synchronous processing, from request to result, using and integrating distributed software components. To exemplify its application, the optimal harvest time for *Brachiaria brizantha* cv. Marandu was determined for the state of Minas Gerais, Brazil, simulating different sowing dates and climate scenarios.

2. Materials and Methods

2.1. Outline of the Model

The WhenHarvest system consist of a simulation model based on the thermal constant of crop development, expressed by the sum of growing degree-days (GDD). The GDD is a climate-based indicator for assessing crop development, which is the sum of heat units when the mean daily temperatures are above the base temperature, below which plant metabolic processes start to cease or dramatically decrease [7]. It is a measure used by farmers to predict the rate of plant development. Despite the complexity of plant growth, this system aims to present a web-based interface that streamlines a widely accepted routine for monitoring plant development and does not consider other plant morphophysiological parameters. It requires the following input parameters such as the latitude and longitude of the location (LAT and LNG, decimal degrees), the desired crop, the sowing date, the soil water holding capacity (WHC, millimeters), and the runoff factor (RF, percentage).

Beginning with latitude and longitude, the system collects a time series of 30 years of climate data from National Aeronautics and Space Administration/Prediction of Worldwide Energy Resources (NASA/POWER; http://power.larc.nasa.gov), at a grid resolution of half a degree of arc of latitude and longitude. Based on the climate data collected, the system predicts the future values of maximum and minimum temperature, and precipitation, using the automated time series forecasting method proposed by Taylor and Letham [8]. Based on the climate data, the system estimates the daily potential evapotranspiration (PET, millimeters) using the Hargreaves equation [9, 10]. The system also determines the actual daily evapotranspiration (AET, millimeters), which in turn depends on the WHC of the soil indicated by the user. Then, with these parameters, the system calculates the water balance for the period, retuning daily values of total soil moisture (ST), water surplus (S), the difference between precipitation (P) and PET, and the runoff (RO). The system pipeline is described in Figure 1.

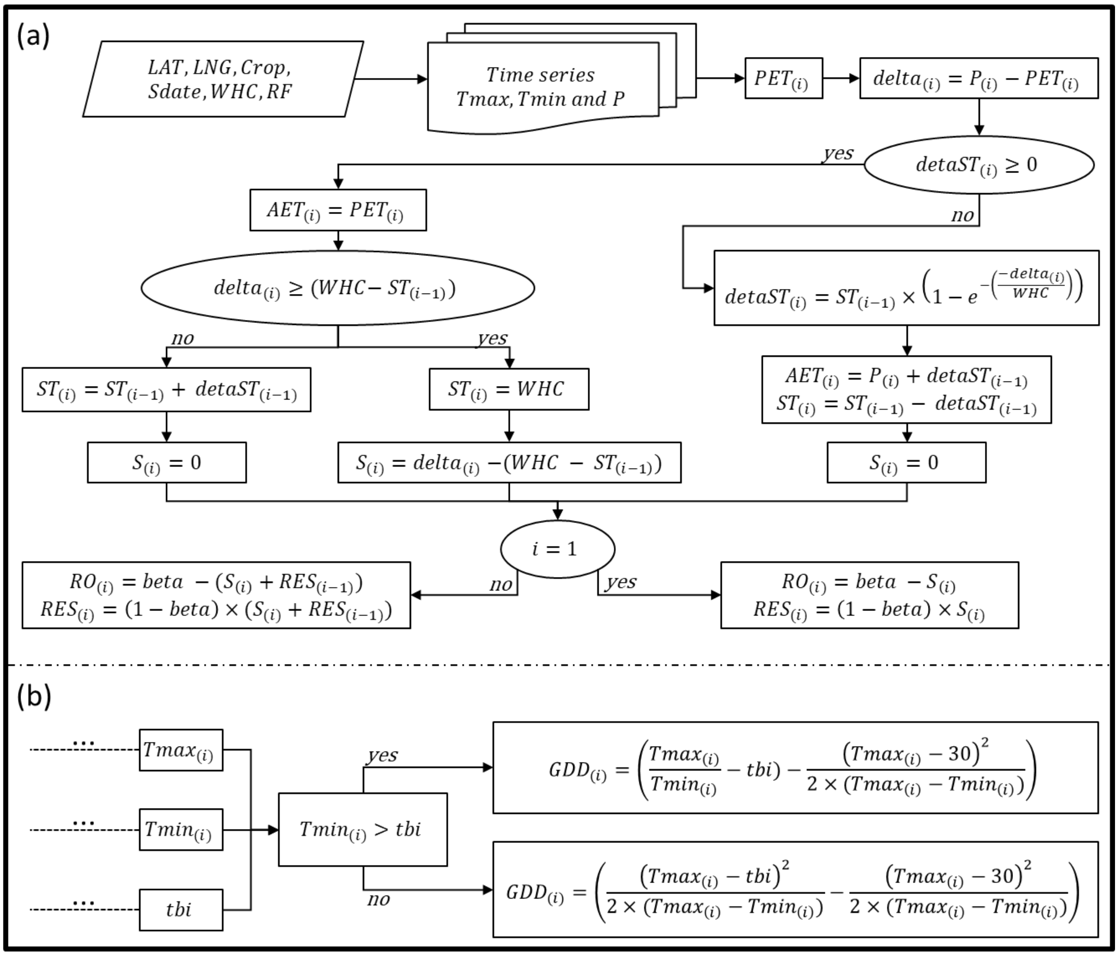


Figure 1. WhenHarvest routine workflow. (a) user input, climate data and water balance; and (b) growing degree-days routine. *LAT* = latitude; *LNG* = longitude; Crop = desired crop; *Sdate* = sowing date; *WHC* = water holding capacity (mm); *RF* = runoff factor (percentage); *Tmax* = maximum temperature (°C); *Tmin* = minimum temperature (°C); *P* = precipitation (mm); *PET* = potential evapotranspiration (mm); delta = *P-PET* (mm); *AET* = actual evapotranspiration (mm); *ST* = soil moisture (mm); *S* = water surplus (mm); *RO* = runoff (mm); $RES_{(i-1)}$ = dynamic water stored in the basin in the previous day (mm); *RES* = dynamic water storage available for the next day (mm); *tbi* = lower base temperature (°C); *GDD* = growing degree-days (°C).

From this interface, it is possible to determine the optimal harvest time in days after sowing (DAS) based on the thermal constant, and obtain information related to the average water balance according to the defined location, considering climatic variations. The scenarios currently provided consider: i) three possibilities of sowing date (considering the sowing date indicated by the user and two variations of ± 14 days); and ii) three climate scenarios (considering the average forecast with uncertainty intervals that represent the best and worst climate scenario with an approximate variation of ± 2°C around the average temperature).

2.2. Exporting Algorithm to the Cloud

The WhenHarvest web-based forecasting system was designed to simplify interaction with users, who are focused on data analysis without installing additional software modules. Therefore, the cloud architecture designed was based on the serverless approach to minimize the complexity of maintaining and scaling the compute. Figure 2 shows the adopted architecture which is based on multi-cloud services.

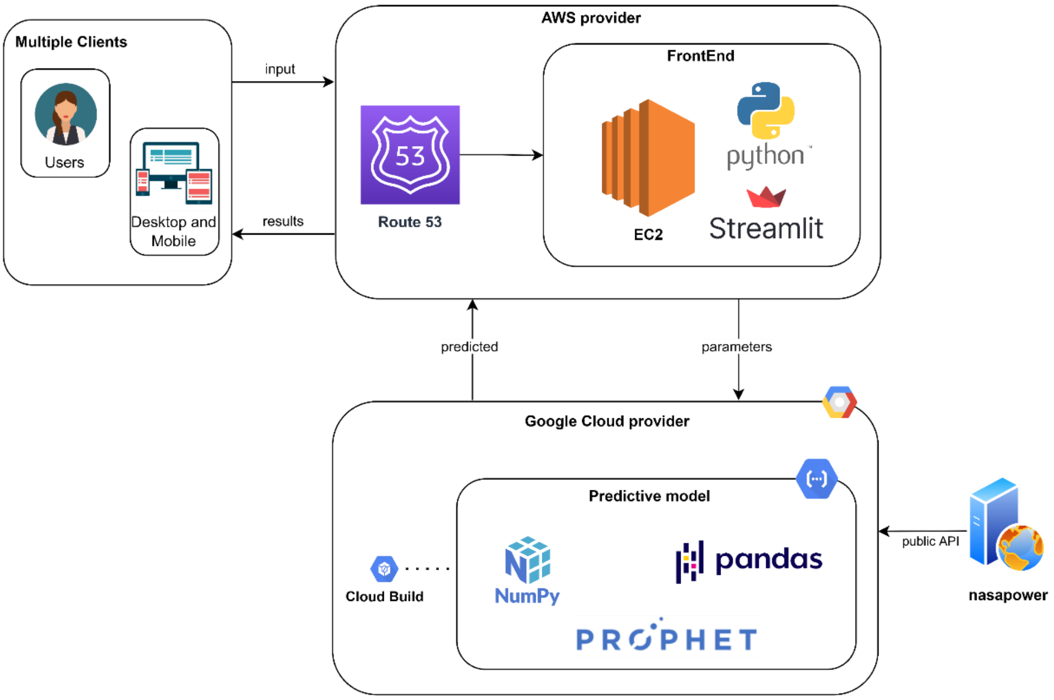
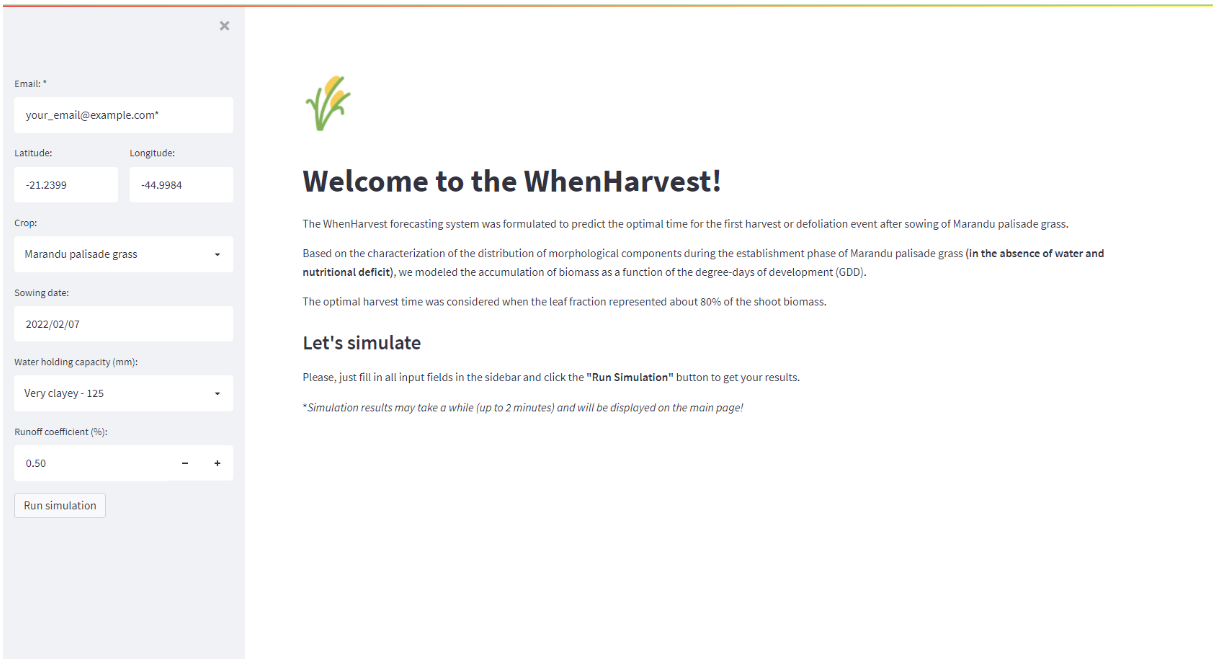


Figure 2. Diagram of the architecture adopted to manage WhenHarvest, a web-based forecast system. The architecture is based on multi-cloud services interconnected with cloud functions to perform the computations.

Currently, the user accesses the WhenHarvest forecast system interface through the website (http://whenharvest.com.br/), which is redirected, via route 53, to the front-end hosted on an EC2 provided by AWS. On the front-end users provide the input information needed for the simulation: (a) E-mail; (b) Latitude; (c) Longitude; (d) Crop; (e) Sowing date; (f) Water holding capacity; and (g) Runoff coefficient. The front end was written using Streamilit, an open-source Python library used to build and deploy data applications (Figure 3). Once the simulation execution is activated, the interface collects, at the same time, triggers the google function where the optimal harvest time predictive model is located. The predictive model was also written in Python using different libraries (Numpy, Pandas and Prophet). At the end of the execution of the predictive algorithm, which takes just a few minutes, the results are automatically displayed on the front-end and are displayed clearly and objectively through graphics (Figures 4 and 5). This approach was used considering the user's ability to define different scenarios and receive the results in real time, making the study, planning and decision-making process agile. Using this architectural framework, we were able to bring together the best of the technology available in the cloud from each of the providers.



Email: *
your_email@example.com*
Latitude: -21.2399
Longitude: -44.9984
Crop: Marandu palisade grass
Sowing date: 2022/02/07
Water holding capacity (mm): Very clayey - 125
Runoff coefficient (%): 0.50

Welcome to the WhenHarvest!

The WhenHarvest forecasting system was formulated to predict the optimal time for the first harvest or defoliation event after sowing of Marandu palisade grass.

Based on the characterization of the distribution of morphological components during the establishment phase of Marandu palisade grass (**in the absence of water and nutritional deficit**), we modeled the accumulation of biomass as a function of the degree-days of development (GDD).

The optimal harvest time was considered when the leaf fraction represented about 80% of the shoot biomass.

Let's simulate

Please, just fill in all input fields in the sidebar and click the **"Run Simulation"** button to get your results.

**Simulation results may take a while (up to 2 minutes) and will be displayed on the main page!*

Figure 3. Developed front-end interface to interact with the end-user to specify the input parameters and run the simulations [http://whenharvest.com.br/, accessed on February 14, 2022].

Figure 4. Example of the optimal harvest time forecast output graph. Sowing dates are plotted against the total number of days (DAS) required for optimal harvest time (closed symbols). The bars before the closed symbols represent the optimal harvest time in the best climate scenario (average T° C +2 °C), while the bars after the symbols represent the optimal harvest time in the worst climate scenario (average T° C -2 °C).

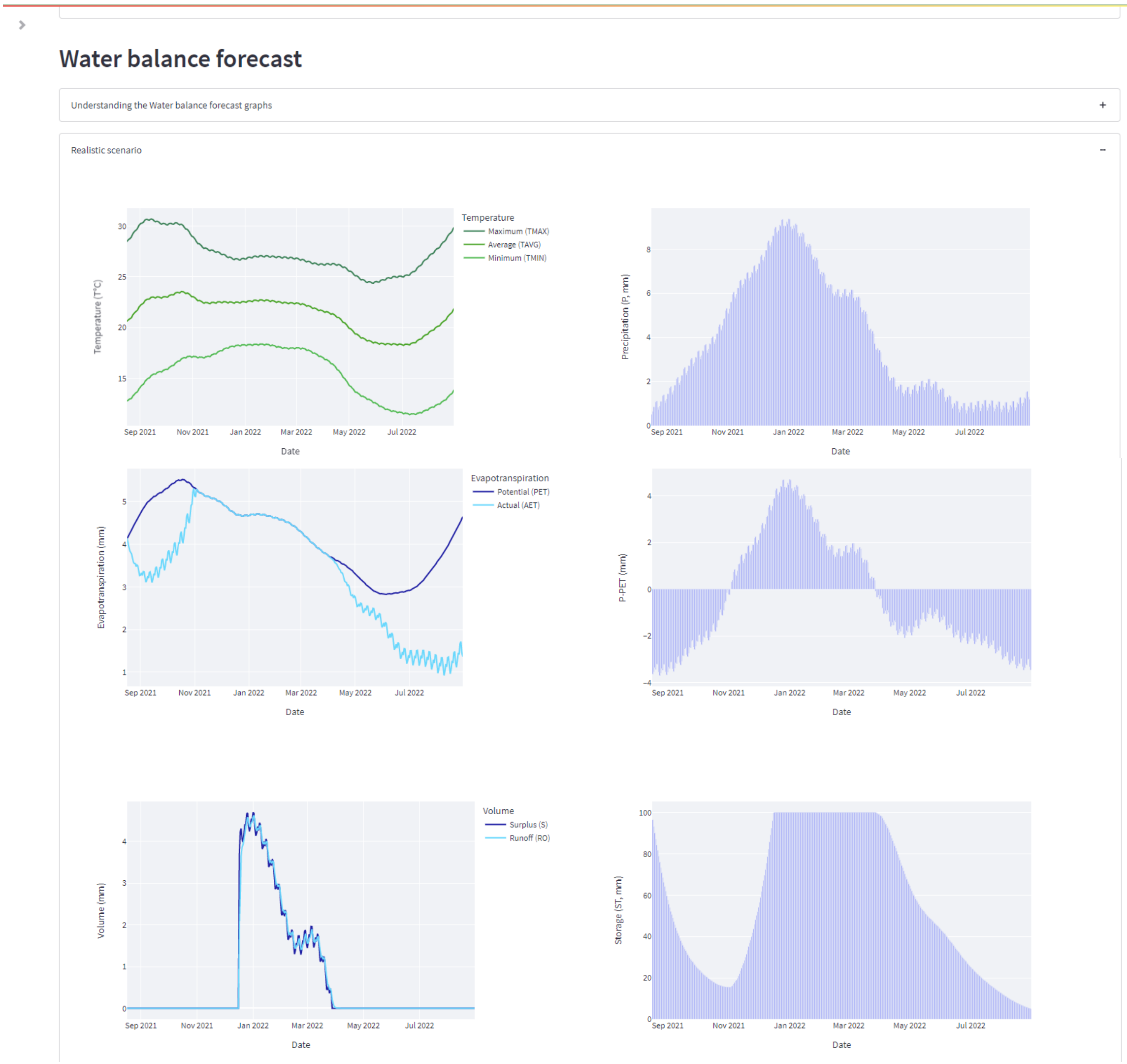


Figure 5. Example of the water balance forecast output graph. Water balance variables are plotted against the date.

2.3. A case study with Marandu palisade grass

From the analysis of plant growth (in the absence of water and nutritional deficit) and the distribution pattern of morphological components over time, it was possible to determine the optimal time for the first harvest or grazing of *Brachiaria brizantha* cv. Marandu after sowing. See Cunha [11] for further details about this experiment. From this study we obtained the necessary parameters to determine the optimal harvest time based on the GDD. To demonstrate the usability of the When Harvest system application, an evaluation was conducted in two phases.

The first phase evaluated the optimal harvest time of Marandu palisade grass simulating sowing on the first day of each month of the year (from January to December 2021), in three distinct locations of the state of Minas Gerais, Brazil. Juvenília located in the extreme north of the state (14°15' S, 44°09' W), the city of Curvelo located in the center of the state (18°46' S, 44° 25' W), and Extrema located in the extreme south of the state (22°51' S, 46°19' W). The second phase expanded the first study by applying the model to all 853 cities in the state of Minas Gerais, simulating sowing between the months of August and March, using a 10-year daily climatological data series (from January 01, 2021, to January 31, 2021).

In both phases, the optimal harvest time was simulated in the different climate scenarios previously described. The best scenario, favorable to plant growth with an increase of up to 2 °C in daily average temperature and the worst scenario, considered less favorable, with a reduction of up to 2 °C in daily average temperature. The simulations were performed considering the WHC parameter equal to 100 mm and the RF parameter equal to 50%. After simulating the optimal harvest time of Marandu palisade grass after sowing, the data were analyzed regarding the structure of the semivariogram structure, passing through the adjustment of spatial dependence and kriging for interpolation and spatial representation of the results. The analyzes and adjustments were performed using the geostatistical analysis functions of the geoR package, of the R statistical program (R CORE TEAM, 2017). The zoning maps were made using ArcMap software version 10.5 (Esri Inc., 2016).

3. Results and Discussion

The results of the first evaluation phase of the web-based forecasting system to determine the optimal harvest time for Marandu palisadegrass were compared for distinct locations over the year, simulating the sow at the beginning of each month (from January to December 2021). The cities in the state of Minas Gerais chosen for the simulation present an approximate difference of 8° of latitude. The locations evaluated were chosen to demonstrate the contrast of latitude and its influences on the GDD accumulation and consequently on the time required to conduct the harvest of Marandu palisade grass after sowing (Figure 6).

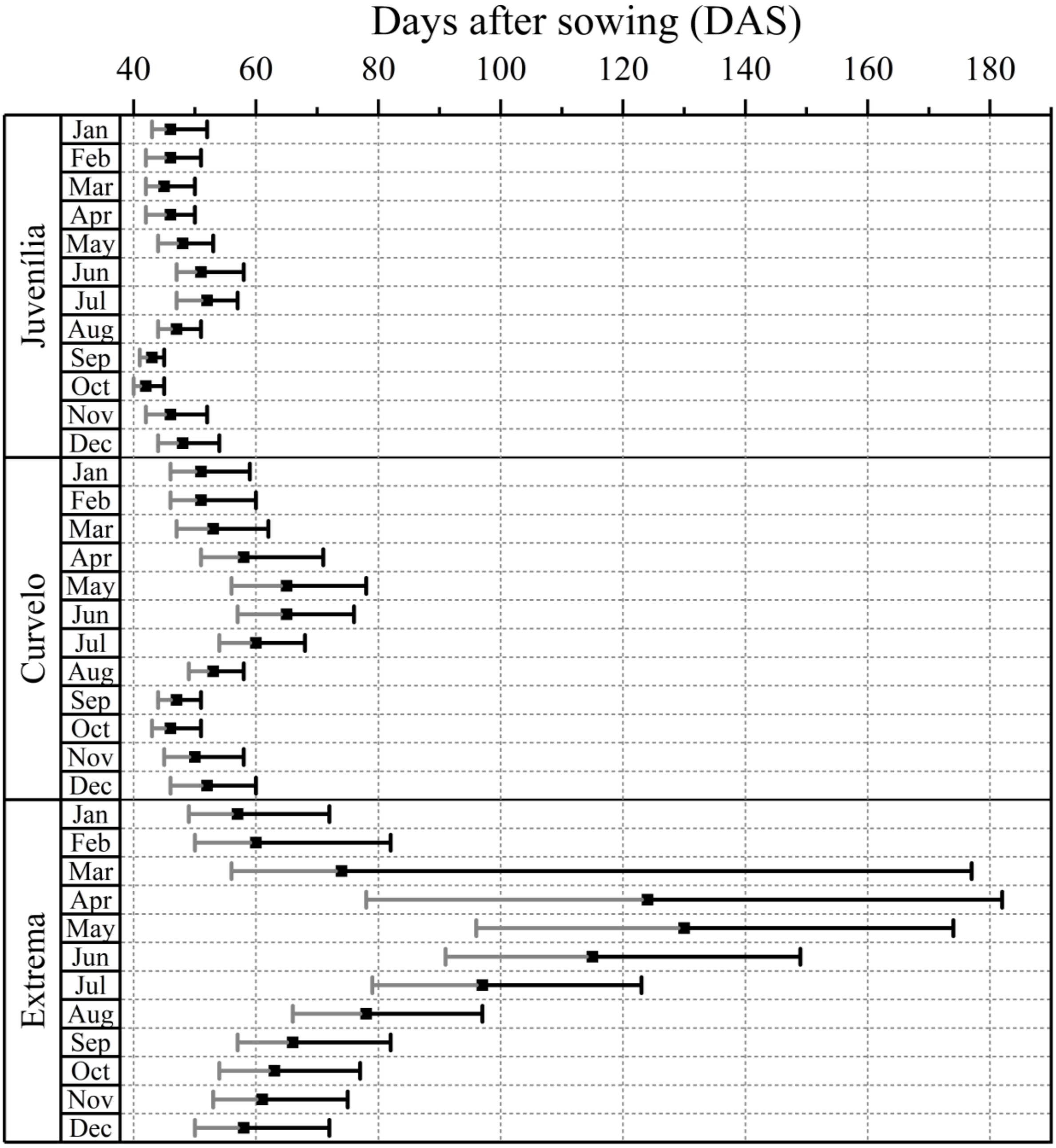


Figure 6. Optimal time to harvest Maradu palisade grass after simulated sowing throughout the year (from January to December 2021), for three different cities in the north, center and south of the state of Minas Gerais, Brazil. The closed squares indicate the optimal harvest time while the gray bars simulate the best climate scenario (average T°C +2°C), and the black bars simulate the worst climate scenario (average T°C -2°C).

As the crop forecasting system is currently based only on the thermal sum and GDD accumulation, the effect of location, latitude and consequently temperature was reflected in the sum of days required for harvest. Juvenília, located in the extreme north of the state, presented a reduced optimal harvest time, with an annual average of 47 DAS, with an average deviation of 3 days considering the best and 5 days considering the worst climate scenario. While Curvelo, located in the center of the state, presented an annual average of 54 DAS, with an average deviation of 6 days considering the best and 8 days considering the worst climate scenario. And Extrema, located in the extreme south of the state, had the longest optimal harvest time, with an annual average of 82 DAS, with an average deviation of 17 days considering the best and 32 days considering the worst climate scenario. These results indicate the magnitude of the effect of location and temperature on plant growth and development. We can also infer that for each city there is a more favorable season for sowing Marandu palisade grass. Considering the month-to-month simulations (Figure 6), we can observe that the most recommended months for sowing in each of the evaluated cities coincide with a reduced deviation of probable days for harvest, comparing the best and the worst climate scenarios. Based just on the temperature criteria, for both Juvenília and Curvelo, sowing was recommended for the months of September and October, while for Extrema, sowing was recommended for December and January. According to the simulations, in these months, there was a high probability of successful harvesting of Marandu palisade grass in a shorter time.

Performing the first harvest or grazing event on newly planted pastures at the optimum harvest point indicates maximizing the use of forage resources. In addition to conduct the harvest in an abbreviated period, this guarantees the vigor and success of the implantation of pastures, promotes the standardization of the canopy, and stimulates tillering. Current literature recommends performing the first grazing 40 to 70 days after germination, when the plants reach 80% of the recommended height. The practical recommendation for conducting the first harvest in pastures is fragile, being commonly expressed in days. In many other cases, the criteria is just based on the observation of the inflorescences followed by the fall of the seeds to the ground, assuming that the deposition of new seeds maximizes the forage production and guarantees the success of the implantation of the pasture. In contrast, the growth analysis performed by Cunha [11], shows that the optimal harvest time occurs before flowering and seed maturation. The optimal moment to conduct the first harvest must consider the morphological aspects of the plants and, at the same time, the influence of the decapitation of the apical meristems on the basal tillering of the plants and the increase in the vigor of the pastures. Cunha [11] considered that the optimal time to harvest Marandu palisade grass after sowing occurred when the leaf fraction represented about 80% of the shoot biomass. Plant growth phase with high leaf area, ideal for ensuring good tiller regrowth and at the same time the high nutritional value of pastures.

Therefore, with the objective of providing subsidies for decision making that help in the successful implementation of pastures, we developed this system to assist in the good planning of agricultural activity. And as a complement to the information on the best moment to sow and harvest, the system also provides the water balance referring to the average climate scenario for the period under study. As the system is based on the thermal sum and the GDD, providing information on the water balance forecast further strengthens the decision-making capacity of farmers. It is a set of complementary information that must be used to assist in the decision of when to sow, evaluating the risks and damages related to the loss of the productive potential of the plants related to the deficit and water stress. Based on input information such as WHC and RF, in addition to variables related to temperature and precipitation, the system calculates the water balance and returns the most important variables (Figure 7) that help in the decision of the sowing plan, which consequently impacts on the best harvest time.

Figure 7. Water balance output graphs from January to December 2021, considering the average climate scenario, in Curvelo, MG, Brazil.

The main results that can be extracted from these output graphs referring to the water balance of Curvelo, MG, calculated for the year 2021 (Figure 7), are related to the dry period between the months from May to October. At the same time, analyzing the differences between P-PET, we identified that the period in which the potential for soil water loss expressed by PET exceeds the entry of water into the soil by rainfall, from April to November. In addition, the soil water storage capacity (*S*) is almost null between September and November. In addition, we can observe the temperature distribution at the study site and its effects on the reduction of GDD accumulation, mainly for the period of the year between May and September. This information, when correlated with the time required to conduct the harvest, helps in the decision-making of the best moment to the sowing of Marandu palisade grass. The association between the necessary period of plant development until the optimal harvest time, with the climate scenario and water availability, allows us to define in a more efficient and precise way the most appropriate moment to conduct the implementation of pastures.

The results of the second evaluation phase of the system for forecasting the optimal harvest time for Marandu palisade grass applied to all cities in Minas Gerais can be seen in Figure 8. The figure is composed of different zoning maps considering the simulation of scenarios with variation of the sowing date (from August to March) and variations of the 10-year climate database with variations around the average (from ±2°C), simulating the best and worst climate scenarios.

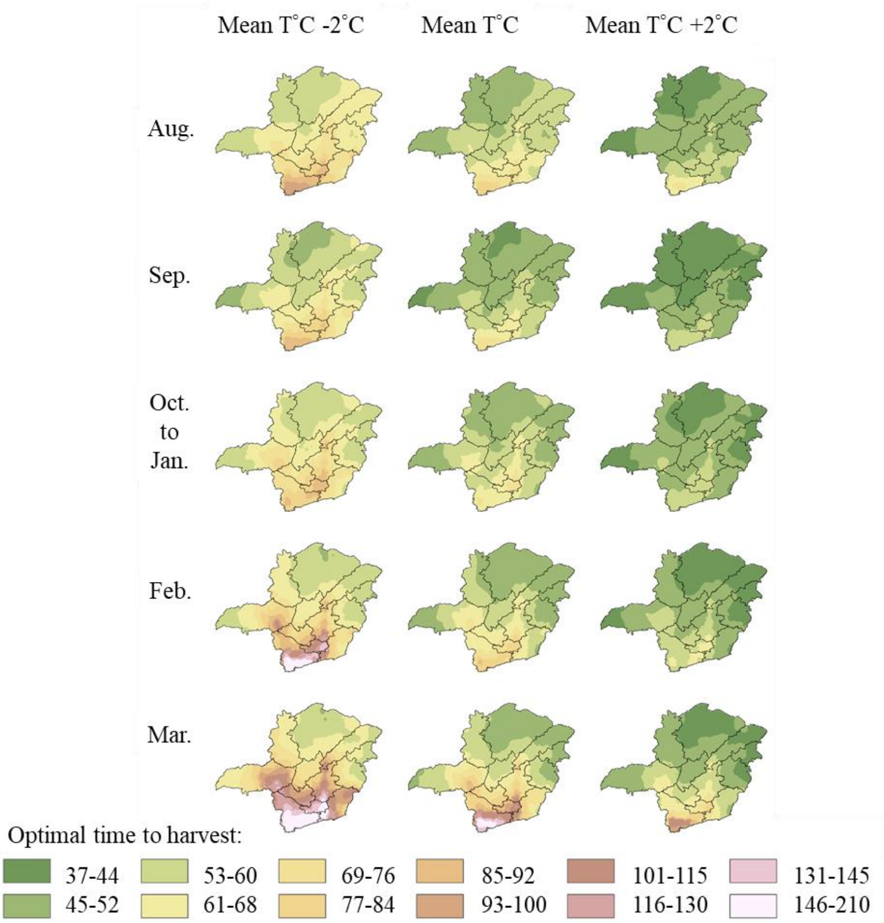


Figure 8. Zoning maps of the optimal time to harvest Marandu palisade grass after simulating sowing simulation between August and March, considering different climate scenarios for the state of Minas Gerais, Brazil.

Evaluating the zoning maps in each of the conditions, we could clearly evidence the effect of the variation of latitude and ambient temperature along the North-South axis for the optimal harvest time of Marandu palisade grass sown in different months of the year. Higher latitudes associated with higher temperatures favor the accumulation of GDD and reduce the chronological time needed to harvest. While in the southern region of the state, when the simulated sowing dates advance to the months of February and March, there is a reduction in the ambient temperature and, consequently, more days are needed to reach the same thermal sum to conduct the harvest.

When we evaluate the most favorable and least favorable climate scenarios for plant growth, this effect becomes even more evident. This evaluation demonstrates that, based only on temperature, the ideal time for sowing and consequently for the first harvest or grazing of pastures formed by Marandu palisade grass can occur at different times for each location. In some regions, such as the center-north of the state, there is greater flexibility and opportunity (larger window) to implement pastures, although limited by the effect of water availability in the system. The regions located in the center-south of the state, on the other hand, have less flexibility (smaller window), being limited mainly by the temperature.

4. Conclusions

The implementation of a web-based forecasting system to determine the optimal harvest time for Marandu palisade grass and the water balance allows users to analyze and plan the sowing and the harvest considering different conditions and scenarios more efficiently, requiring simple input variables for simulation. End-users will have the necessary instructions for using the system, as well as general explanations for the good use of the information generated. Data (input and output) is managed through multiple cloud services, decoupling each component, and allowing automatic scaling, according to the number of requests. The results are shown after the simulations on the same page where the input data is entered, allowing the process to be run multiple times using different input parameters, in just a few minutes. With this system, it is possible to assess the success of a sowing and its impacts more quickly at the time of harvest in different contexts and discuss the advantages and disadvantages associated with each simulation scenario. In addition, there is no need to install additional software modules, which simplifies interaction with the end user, who can concentrate on analyzing and interpreting the results. Future improvements of the WhenHarvest, may involve: i) the acquisition of meteorological data from real meteorological stations in the national territory; ii) improvement of rain forecast models; iii) the addition of a factor that simulates the effect of water stress on plant growth; iv) the inclusion of other methods for determining potential evapotranspiration; v) the inclusion of other crops in the forecast system; and vi) the inclusion of other important phenological stages in the growth and development of plants.

Author Contributions

Conceptualization, D.C. and M.L.; methodology, D.C.; software, D.C., M.O. and M.M.; validation, D.C. and M.L.; writing – original draft preparation, D.C.; writing – review and editing, all authors.

Conflicts of Interest

The authors declare no conflict of interest.

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