

## Short Communication

# The effect of stage of regrowth on the physical composition and nutritive value of the various vertical strata of kikuyu (*Cenchrus clandestinus*) pastures

*Efecto de la edad de rebrote en la composición física y el valor nutritivo de los diferentes estratos verticales de pasturas de kikuyo (Cenchrus clandestinus)*

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## Abstract

A plot study was conducted at the Gatton Research Dairy, Queensland, Australia, to quantify the effects of 5 regrowth periods (9, 11, 14, 16 and 18 days) and 4 vertical strata on the composition and nutritive value of kikuyu (*Cenchrus clandestinus*) pastures using a block factorial design with 4 replicates. Pasture samples were analyzed for crude protein (CP), ethanol-soluble carbohydrates (ESC), acid detergent fiber (ADF), neutral detergent fiber (aNDFom), in vitro indigestible neutral detergent fibre (iNDF240) and minerals. Metabolizable energy (ME) was then calculated from the concentrations of other nutrients. Regardless of the stage of regrowth, stems were located mainly in the bottom 1 or 2 strata, while leaves were present mainly in the top 2 or 3 strata. CP, ESC and ME declined, but aNDFom, ADF and iNDF240 increased with stage of regrowth and from top to bottom of the swards ( $P < 0.05$ ). While herbage quality variables were affected by both factors, vertical stratum had a much larger impact on quality than stage of regrowth. These results indicate that grazing management of kikuyu pastures should be based not only on stage of regrowth but also on level of defoliation, as both have strong impacts on the nutritive value of the consumed forage.

**Keywords:** Chemical composition, grazing management, leaf stage, sward structure, tropical pastures.

## Resumen

En Gatton Research Dairy, Queensland, Australia, en un diseño factorial en bloques con cuatro repeticiones, se evaluó el efecto de cinco periodos de rebrote (9, 11, 14, 16 y 18 días) y cuatro estratos verticales en el valor nutritivo de pasturas de kikuyo (*Cenchrus clandestinus*). En muestras de pasturas se determinaron la concentración de proteína cruda (PC), carbohidratos solubles en etanol (CSE), fibra detergente ácido (FDA) y neutro (FDN), FDN indigestible (FDNi), energía metabolizable (EM) y minerales. Los resultados mostraron que los tallos estuvieron localizados principalmente en los dos estratos inferiores y las hojas en los dos o tres estratos superiores de las pasturas. Consecuentemente, la PC, CSE y EM se redujeron, y FDA, FDN y FDNi se incrementaron con el estado de rebrote y desde la parte superior a la inferior de la pastura. A pesar de que ambos factores experimentales afectaron la calidad del forraje, los estratos verticales afectaron más a la calidad que el estado de rebrote. Estos resultados indican que los dos factores deben ser considerados para el manejo del kikuyo ya que ambos afectan significativamente a la calidad del forraje ingerido.

**Palabras clave:** Composición química, estructura de pasto, manejo de pastoreo, pasturas tropicales.

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## Introduction

Kikuyu (*Cenchrus clandestinus* syn. *Pennisetum clandestinum*) is one of the main improved pasture species used by the grazing industries in the northeast of Australia and other tropical and subtropical areas of the world. Previous studies examined the effects of stage of regrowth on herbage quality of leaves, stems and whole plants of kikuyu pastures (Reeves et al. 1996). These early studies provided essential data for the understanding of factors driving herbage quality in kikuyu, such as differences in quality between plant parts at different stages of regrowth; however, these herbage quality results may not necessarily represent the quality of herbage actually consumed by animals grazing these pastures.

It is well known from previous studies that beef and dairy cattle graze pastures in strata, and herbage quality declines from the top to the bottom of the swards (Ungar and Ravid 1999; Benvenuti et al. 2016, 2017; Ison et al. 2018). These studies found that, when grazing pressure increased, cattle were forced to graze forage in the lower strata which was of lower quality and as a result diet quality and animal performance declined. It is important to quantify the herbage quality of different strata of kikuyu pastures to assist members of the grazing industry in making informed decisions on grazing management, based on a sound understanding of the effects of sward structure on diet quality. The aim of this study was to quantify the effects of stage of regrowth and vertical distribution in the sward on physical composition and nutritive value of kikuyu pastures.

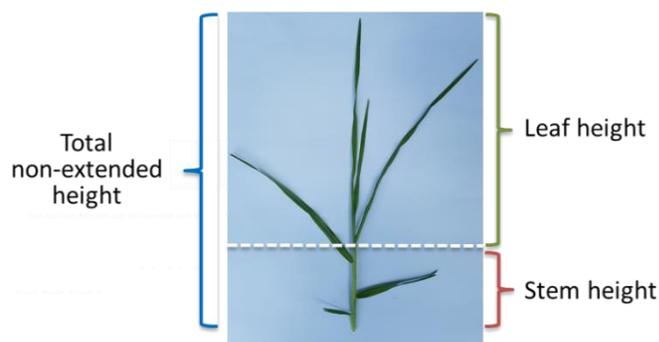
## Materials and Methods

### *Experimental design and pasture assessment*

The study was conducted during 18 days on established kikuyu pastures at the Gatton Research Dairy, Queensland, Australia (27°32'45" S, 152°19'44" E; 104 masl) in January–February 2016. The kikuyu pasture was planted in 2010 into a black alluvial soil. The effects of 5 regrowth periods (9, 11, 14, 16 and 18 days) and 4 vertical strata on physical structure and nutritive value of the pasture were assessed using a blocked factorial design with 4 replicates. On 18 January 2016 the experimental area (0.2 ha) of kikuyu pasture was divided into 4 blocks of 0.05 ha each and mown to 5 cm in height before 100 kg urea/ha was applied. Plots of 9 m<sup>2</sup> within blocks were randomly allocated to differing regrowth periods. The pasture was irrigated regularly to replace evapotranspiration losses. During the 3 weeks of the experimental period 44 mm of rainfall was received and 65 mm

irrigation was applied. A single 1 m<sup>2</sup> sampling area was used on each occasion to assess physical attributes and perform harvests to determine yield and quality attributes of pasture in each of the 4 blocks for the 5 regrowth periods.

Prior to harvesting the 1 m<sup>2</sup> sampling area, 30 random tillers were measured for total non-extended height, stem height and number of fully-expanded leaves (Figure 1). Stem height was defined as the height from ground level to the base of the lamina (ligula) of the top fully-expanded leaf. The averages of total sward height, stem height and number of leaves were then calculated.



**Figure 1.** Heights of tillers and plant parts.

Pasture inside the sampling quadrat was then cut in 4 vertical strata. The strata were numbered from top to bottom of the sward so that stratum 1 was the top stratum. The bottom stratum consisted of plant material collected from ground level to 5 cm high. The top 3 strata were equal vertical proportions of pasture above 5 cm. Average total sward height minus 5 cm was divided by 3 to form the 3 strata, indicating that the depth of each stratum varied with the total height of the sward. Within each stratum, harvested material was dried at 60 °C to determine dry matter (DM) yield. Samples from each stratum were combined in pairs of replicates (blocks 1 and 2 and blocks 3 and 4). Combined subsamples from each stratum were then sent for analysis to the Dairy One Forage Lab (Ithaca, NY, USA) to determine crude protein (CP), ethanol-soluble carbohydrates (ESC), acid detergent fiber (ADF), amylase, sodium sulphite-treated neutral detergent fiber expressed on an ash-free basis (aNDFom) and minerals. Subsamples of each stratum were also analyzed for in vitro indigestible NDF from 240-hour incubations with rumen fluid (iNDF240). Metabolizable energy (ME) values were calculated using equations and relationships with other nutrients. The Dairy One Forage Lab uses a multiple component summative approach for its ruminant energy prediction system:

ME (MJ/kg DM) =  $(1.01 \times 0.04409 \times \text{TDN} - 0.45) \times 4.184$ ,

where: TDN is total digestible nutrients (%).

#### Chemical analysis and in vitro rumen fermentation

The ADF and aNDFom concentrations were determined using the ANKOM Model 200 and the fiber bag technique developed by ANKOM (ANKOM Technology, Macedon, NY, USA). The acid and neutral solutions for these analyses were prepared as per AOAC 973.18 (AOAC 1984) and Van Soest et al. (1991), respectively. ESC was determined by the method of Hall et al. (1999). Samples were analyzed for CP using the AOAC procedure 990.03 (AOAC 1984). To determine mineral concentrations, samples were digested using CEM Microwave Accelerated Reaction System (MARS6) with MarsXpress Temperature Control using 50 ml calibrated Xpress Teflon PFA vessels with Kevlar/fiberglass insulating sleeves then analyzed by ICP using a Thermo iCAP 6300 Inductively Coupled Plasma Radial Spectrometer.

The iNDF240 concentration was determined using 240-hour in vitro fermentation in Daisy Incubators (ANKOM Technology, Macedon, NY, USA) set at 39 °C. Each Daisy incubation cupboard can incubate 4 bottles (24 bags of samples per bottle) at a time with 1,520 mL buffer solution added to each bottle and then combined with 400 mL rumen fluid taken from a fistulated steer. After 120 h fresh rumen fluid was collected and solutions were replenished. After 240 h, bags were removed and rinsed until clear. NDF concentration in the residue was analyzed using the Ankom Fiber Analyzer.

#### Statistical analysis

Nutritive values for herbage components were analyzed according to a blocked 5 × 4 factorial design using analysis of variance. Here the fixed effects were stage of regrowth at the main-plot level, and vertical stratum as a split-plot effect. GenStat (2016) was used for the analyses.

## Results and Discussion

#### Pasture structure

Leaf numbers in the pastures ranged from 2.2 to 4.7 leaves per tiller (Table 1). This range included the currently recommended stage of regrowth of 4.5 leaves per tiller for grazing kikuyu pastures (Reeves et al. 1996). Regardless of stage of regrowth, all pastures consisted of the same general structure, with stems predominantly located at the bottom of the sward and leaves at the top of the sward (Table 1). The average height of the stems, as a proportion of total sward height, increased from 0.32 to 0.44 as stage of growth advanced from 1 to 5 (Table 1). This indicated that the proportion of stems increased from top to bottom of the sward as well as with the stage of regrowth. A large proportion of the bottom 1 or 2 vertical strata consisted of stems and the top 2 or 3 strata consisted mainly of leaves (Table 2). Similarly, Benvenuti et al. (2016) found that pastures of *Axonopus catarinensis* had a similar sward structure, where the stems were located in the bottom one-third of the sward for a range of regrowth stages. The results are also consistent with the study by Reeves et al. (1996), who found that the proportion of stems increased with the stage of regrowth of kikuyu pastures.

**Table 1.** Physical structure of kikuyu swards at different stages of regrowth. See also Figure 1.

| Stage of regrowth<br>(no. of days) | Pasture height<br>(cm) | Stem height<br>(cm) | Stem height (proportion<br>of pasture height) | Leaf height<br>(cm) | Fully expanded<br>leaves (no.) |
|------------------------------------|------------------------|---------------------|-----------------------------------------------|---------------------|--------------------------------|
| 1 (9)                              | 13.9                   | 4.4                 | 0.32                                          | 9.5                 | 2.2                            |
| 2 (11)                             | 16.3                   | 5.3                 | 0.32                                          | 11.0                | 2.5                            |
| 3 (14)                             | 23.7                   | 9.1                 | 0.38                                          | 14.6                | 3.3                            |
| 4 (16)                             | 29.1                   | 12.5                | 0.43                                          | 16.5                | 4.1                            |
| 5 (18)                             | 35.1                   | 15.6                | 0.44                                          | 19.5                | 4.7                            |
| Probability                        | <0.001                 | <0.001              | <0.001                                        | <0.001              | <0.001                         |
| s.e.m.                             | 0.798                  | 0.563               | 0.011                                         | 0.423               | 0.068                          |
| LSD (P<0.05)                       | 2.46                   | 1.74                | 0.033                                         | 1.30                | 0.21                           |

**Table 2.** The effects of stage of regrowth (SoR) on stem proportion and herbage quality (DM basis) of vertical strata (S1–S4) of kikuyu pastures. S1 to S4: top to bottom stratum.

| Regrowth stage and stratum | Stem height (proportion of stratum height) | Crude protein (g/kg) | ADF (g/kg) | aNDFom (g/kg) | iNDF240 (g/kg) | ESC (g/kg) | ME (MJ/kg) | Calcium (g/kg) | Phosphorus (g/kg) | Potassium (g/kg) | Sodium (g/kg) |
|----------------------------|--------------------------------------------|----------------------|------------|---------------|----------------|------------|------------|----------------|-------------------|------------------|---------------|
| SoR 1 (9 days)             |                                            |                      |            |               |                |            |            |                |                   |                  |               |
| S1                         | 0.0                                        | 340                  | 212        | 371           | 0.05           | 68.5       | 11.8       | 4.60           | 4.60              | 29.9             | 1.35          |
| S2                         | 0.0                                        | 338                  | 252        | 375           | 0.05           | 54.5       | 10.8       | 4.35           | 4.90              | 35.6             | 1.58          |
| S3                         | 0.0                                        | 310                  | 277        | 445           | 0.08           | 46.0       | 9.8        | 4.85           | 5.10              | 38.3             | 1.86          |
| S4                         | 0.8                                        | 255                  | 333        | 561           | 0.21           | 37.5       | 8.7        | 6.90           | 4.55              | 36.7             | 2.46          |
| SoR 2 (11 days)            |                                            |                      |            |               |                |            |            |                |                   |                  |               |
| S1                         | 0.0                                        | 357                  | 228        | 417           | 0.07           | 32.5       | 11.9       | 4.35           | 5.20              | 39.7             | 1.25          |
| S2                         | 0.0                                        | 335                  | 300        | 462           | 0.08           | 30.0       | 9.5        | 4.10           | 5.70              | 46.6             | 1.61          |
| S3                         | 0.0                                        | 302                  | 352        | 512           | 0.10           | 26.0       | 8.6        | 4.90           | 5.90              | 49.5             | 1.95          |
| S4                         | 0.9                                        | 247                  | 366        | 595           | 0.24           | 34.0       | 8.0        | 7.50           | 4.65              | 37.2             | 2.29          |
| SoR 3 (14 days)            |                                            |                      |            |               |                |            |            |                |                   |                  |               |
| S1                         | 0.0                                        | 350                  | 294        | 422           | 0.06           | 39.0       | 10.7       | 3.90           | 5.25              | 40.7             | 0.94          |
| S2                         | 0.0                                        | 314                  | 319        | 462           | 0.09           | 22.0       | 9.4        | 3.35           | 6.00              | 51.4             | 1.16          |
| S3                         | 0.5                                        | 295                  | 309        | 495           | 0.13           | 18.0       | 9.5        | 3.85           | 6.05              | 54.9             | 1.52          |
| S4                         | 1.0                                        | 242                  | 352        | 600           | 0.28           | 28.0       | 8.4        | 6.80           | 5.00              | 41.5             | 2.20          |
| SoR 4 (16 days)            |                                            |                      |            |               |                |            |            |                |                   |                  |               |
| S1                         | 0.0                                        | 329                  | 287        | 446           | 0.07           | 39.5       | 9.6        | 3.90           | 4.60              | 45.1             | 0.75          |
| S2                         | 0.0                                        | 303                  | 280        | 491           | 0.08           | 31.5       | 9.8        | 3.05           | 5.15              | 55.3             | 0.98          |
| S3                         | 0.9                                        | 279                  | 322        | 510           | 0.13           | 26.0       | 8.9        | 3.90           | 5.40              | 59.3             | 1.47          |
| S4                         | 1.0                                        | 228                  | 362        | 602           | 0.28           | 19.0       | 8.0        | 6.75           | 3.95              | 40.7             | 2.14          |
| SoR 5 (18 days)            |                                            |                      |            |               |                |            |            |                |                   |                  |               |
| S1                         | 0.0                                        | 314                  | 285        | 451           | 0.07           | 42.5       | 10.3       | 3.95           | 4.60              | 43.2             | 1.13          |
| S2                         | 0.0                                        | 293                  | 284        | 497           | 0.08           | 37.5       | 10.0       | 3.35           | 5.25              | 54.5             | 1.23          |
| S3                         | 0.9                                        | 268                  | 329        | 522           | 0.13           | 27.5       | 8.8        | 4.15           | 5.05              | 59.2             | 1.60          |
| S4                         | 1.0                                        | 226                  | 369        | 600           | 0.26           | 28.0       | 7.5        | 6.70           | 4.20              | 45.8             | 2.29          |
| P SoR                      | 0.002                                      | 0.049                | 0.072      | 0.014         | <0.001         | 0.15       | 0.009      | 0.008          | 0.029             | 0.01             | <0.001        |
| P Stratum                  | <0.001                                     | <0.001               | <0.001     | <0.001        | <0.001         | 0.01       | <0.001     | <0.001         | <0.001            | <0.001           | <0.001        |
| P SoR × Stratum            | <0.001                                     | 0.046                | 0.068      | 0.032         | 0.104          | 0.735      | 0.004      | 0.389          | <0.001            | <0.001           | 0.053         |
| s.e.m.                     | 0.04                                       | 7.18                 | 15.14      | 11.49         | 0.007          | 8.29       | 0.25       | 0.20           | 0.14              | 1.70             | 0.05          |
| LSD (P<0.05)               | 0.12                                       | 24.1                 | 45.3       | 36            | 0.022          | 25.2       | 0.74       | 0.59           | 0.50              | 6.09             | 0.16          |

ESC – ethanol-soluble carbohydrates; ADF – acid detergent fiber; aNDFom – amylase and sodium sulphite-treated neutral detergent fiber expressed on an ash-free basis; iNDF240 – in vitro indigestible neutral detergent fiber from 240-hour incubations with rumen fluid; ME – metabolizable energy; stratum numbering from top (S1) to bottom (S4).

### Nutritive value

Since there was a significant interaction ( $P<0.05$ ) between experimental factors (stages of regrowth and vertical strata) for most herbage quality variables, results are shown for all combinations of factors in Table 2. While herbage quality was significantly affected by both factors, vertical stratum had a much larger impact on quality than stage of regrowth for most quality variables. This is explained in detail below for each herbage quality variable.

It is likely that the observed differences in herbage quality between vertical strata and between stages of regrowth were due to the differences in the proportion of different plant parts or to their changes in quality as the pasture matured (Table 2). The differences in herbage quality between strata were largely due to the differences

in the proportions of leaves and stems in the different strata (Table 2). Consistently, Fulkerson et al. (2010) reported a significant difference in herbage quality between leaves and stems in kikuyu pastures. Since herbage quality differed between stages of regrowth for all strata, the observed differences in quality between stages of regrowth could be attributed to changes in quality of individual plant parts as the pasture matured. Reeves et al. (1996) found that the CP and mineral concentrations of leaves changed significantly with the stage of regrowth of kikuyu pastures.

CP and ESC concentrations declined significantly as stage of regrowth increased and from top to bottom of the swards ( $P<0.05$ ). These quality variables were more affected by the vertical stratum than the stage of regrowth. While the average decreases in CP and ESC concen-

trations from stage 1 to stage 5 of regrowth were 11 and 34%, respectively, the average declines from the top to the bottom stratum were 25 and 44%, respectively. Previous studies also found that CP% declined with the stage of regrowth in kikuyu pastures (Reeves et al. 1996) and from top to bottom in pastures of *Axonopus catarinensis* (Benvenuti et al. 2016).

In contrast, concentrations of aNDFom, ADF and iNDF240 increased significantly with stage of regrowth and from top to bottom of the sward ( $P < 0.05$ ). These variables were also more affected by the vertical stratum than the stage of regrowth. While the average increases in concentrations of aNDFom, ADF and iNDF240 from stage 1 of regrowth to stage 5 were 20, 19 and 48%, the average increases from the top to the bottom stratum were 40, 36 and 300%, respectively. These results indicate that the digestibility of the plant material would decrease at later stages of regrowth and from top to bottom of the sward. This confirms the work of Reeves et al. (1996), who found that organic matter digestibility decreased as stage of regrowth increased in kikuyu pastures.

As might be expected, ME decreased consistently when ESC decreased and aNDFom, ADF and iNDF240 increased. Therefore, ME significantly declined with stage of regrowth and from top to bottom of the sward ( $P < 0.05$ ). This quality variable was also more affected by the vertical stratum than stage of regrowth. While the average decrease of ME from stage 1 of regrowth to stage 5 was 11%, the average decline from the top to the bottom stratum was 25%.

The effects of stage of regrowth and vertical stratum on mineral concentration in the plant material differed between minerals (Table 2). Concentrations of calcium and sodium increased from top to bottom of the sward, while concentrations of potassium and phosphorus increased from stratum 1 to stratum 3 and then decreased for the bottom stratum. Unlike potassium, concentrations of calcium and sodium decreased with stage of regrowth. In turn, the concentration of phosphorus increased from stage 1 of regrowth to stage 3 and then decreased. On the contrary, Reeves et al. (1996) found that, as leaves of kikuyu pastures matured, calcium concentration increased while those of potassium and phosphorus decreased, but sodium concentration did not change with maturity.

## Conclusion

This study has shown that, regardless of the stage of regrowth, stems were located mainly in the bottom one or two strata, while leaves were present mainly in the top two or three strata. This indicates that herbage quality declines significantly from top to bottom of the sward as it is

grazed. In addition, herbage quality declines significantly with the stage of regrowth for all vertical strata, probably as a result of changes in nutritive value within plant parts as the pasture matures. While herbage quality was affected by both factors, the vertical stratum had a much larger impact on nutritive value than stage of regrowth.

We conclude that grazing management of kikuyu pastures should be based not only on the stage of regrowth but also on the level of defoliation, as both have strong impacts on the herbage quality of forage consumed by the animals. By considering these issues, members of the grazing industry can now exert greater control over the nutritive value of the forage consumed by the animals by controlling the level of defoliation of the pasture according to its stage of regrowth. Similar studies on other pasture species, e.g. erect species, would verify if these concepts are relevant to species with different growth habits.

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