ผลของแหล่งอาหารหยาบ และระบบการให้อาหาร ต่อกระบวนการหมักในรูเมน การย่อยได้ของโภชนะ และคุณภาพเนื้อของแพะ

นางสาวอัจฉรา ลักขณานุกูล

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรดุษฎีบัณฑิต
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EFFECTS OF FORAGE SOURCES AND FEEDING REGIMES ON RUMEN FERMENTATION, NUTRITION DIGESTIBILITY AND GOAT MEAT QUALITY

Achara Lukkananukool

A Thesis Submitted in Partial Fulfillment of the Requirements for the Degree of Doctor of Philosophy in Animal Production Technology

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EFFECTS OF FORAGE SOURCES AND FEEDING REGIMES ON RUMEN FERMENTATION, NUTRITION DIGESTIBILITY AND GOAT MEAT QUALITY

Suranaree University of Technology has approved this thesis submitted in partial fulfillment of the requirements for the Degree of Doctor of Philosophy.

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วิทยานิพนธ์นี้มีวัตถุประสงค์เพื่อศึกษาชนิดของพืชอาหารสัตว์เขตร้อนและรูปแบบการให้ที่เหมาะสมต่อการเลี้ยงแพะเนื้อ การศึกษาครั้งนี้ประกอบด้วย 3 การทดลอง

การทดลองที่ 1 พืชอาหารสัตว์เขตร้อน 6 ชนิด ได้แก่ หญ้ากินนีสีม่วง (PG; *Panicum maximum* TD. 58), หญ้ามูลาโต้ 2 (MG; *Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*), Napiergrass (NG; *Pennisetum purpureum* Schumacher), ถั่วท่าพระสไตโล (TS ; *Stylosanthes guianensis* CIAT 184), ถั่วฮามาต้า (VS; *Stylosanthes hamata*) และถั่วคาวาเคด (CC; *Centrosema pascuorum*) เก็บเกี่ยวที่อายุ 30, 45 และ 60 วันหลังการงอกใหม่ภายหลังการตัด (days after regrowth) พืชอาหารสัตว์ส่วนหนึ่งนำไปทำการหมัก โดยมีการใช้สารเสริม คือ มันเส้นบดกากน้ำตาล น้ำพืชหมัก (FJLB) และไม่ใช้สารเสริม จากผลการศึกษา พบว่า เมื่ออายุการตัดของพืชอาหารสัตว์เพิ่มขึ้น ปริมาณเยื่อใยที่ละลายในสารละลายที่เป็นกลาง (NDF) และเยื่อใยที่ละลายในสารละลายที่เป็นกรด (ADF) เพิ่มขึ้น แต่ปริมาณของอินทรียวัตถุ (OM) โปรตีนหยาบ (CP) และไขมัน (EE) กลับลดลง องค์ประกอบทางเคมีของพืชอาหารสัตว์หมักมีความสัมพันธ์เช่นเดียวกับพืชอาหารสัตว์สด กรดไขมันหลักที่พบในพืชอาหารสัตว์หมักนั้นเหมือนกับที่พบในพืชอาหารสัตว์สด แต่รูปแบบของกรดไขมันมีการเปลี่ยนแปลงไปเล็กน้อย

การทดลองที่ 2 ทำการศึกษาแหล่งอาหารหยาบ อาหารทดลองมี 4 ทรีทเมนต์ คือ 1) หญ้ามูลาโต 2 ในรูปแบบสด 2) ถั่วฮามาตาในรูปแบบสด 3) หญ้ามูลาโต 2 ในรูปแบบหมักด้วย FJLB และ 4) ถั่วฮามาตาในรูปแบบหมักด้วย FJLB ในแพะเจาะกระเพาะเพศผู้ลูกผสมพันธุ์บอร์และแองโกลนูเบียน จำนวน 8 ตัว ใช้แผนการทดลองแบบ 2 × 2 factorial arrangements in 4 × 4 replicated Latin square จากผลการศึกษา พบว่า อาหารทดลองทุกทรีทเมนต์มีผลต่อกระบวนการหมักในรูเมน (Rumen fermentation) ไม่แตกต่างการทางสถิติ (P>0.05) กรดไขมันที่พบเป็นองค์ประกอบหลักของน้ำรูเมน (Rumen fluid) คือ C18:0 และ C18:1n9 ซึ่งถูกเปลี่ยนมาจากกรดไขมัน C18:2n6 และ C18:3n3 โดยจุลินทรีย์ในกระเพาะรูเมนด้วยกระบวนการ biohydrogenation

การทดลองที่ 3 ทำการศึกษาแหล่งและรูปแบบการให้อาหารหยาบในแพะ โดยใช้พืชอาหารสัตว์ 2 ชนิด คือ หญ้ามูลาโต้ 2 และ ถั่วฮามาตา รูปแบบการให้อาหารหยาบมี 3 แบบ คือ 1) แบบตัดสดและนำไปให้กิน 2) แบบพืชอาหารสัตว์หมัก และ 3) แบบปล่อยแทะเล็ม ใช้แพะเนื้อเพศผู้

ลูกผสมพันธุ์บอร์และแองโกลนูเบียน จำนวน 30 ตัว จัดกลุ่มการทดลองโดยใช้ 2 × 3 factorial arrangements in CRD จากผลการศึกษา พบว่า แพะในกลุ่มที่ปล่อยแทะเล็มถั่วฮามาตามีปริมาณการกินได้สูงสุด (P<0.01) แพะที่ปล่อยแทะเล็มมีเปอร์เซ็นต์ซาก (dressing percentage) ต่ำ และค่าความเป็นกรด-ด่างภายหลังการฆ่าต่ำ แต่พื้นที่หน้าตัดเนื้อสัน เปอร์เซ็นต์เนื้อแดง (% lean) และการสูญเสียน้ำ (% drip loss) มากกว่าแพะในกลุ่มอื่นๆ แพะที่ได้รับพืชอาหารสัตว์ทั้งสองชนิดในรูปแบบของการหมักมีปริมาณ conjugated linoleic acid (CLA) ในเนื้อสันนอกต่ำที่สุด (P<0.05) สัดส่วนของกรดไขมัน n-6 / n-3 ในกล้ามเนื้อสันนอกในแพะที่ได้รับพืชอาหารสัตว์แบบตัดสดแล้วนำไปให้กิน และแบบปล่อยแทะเล็มมีค่าอยู่ในช่วง 3.33-4.52 ดังนั้น การให้พืชอาหารสัตว์สองชนิดนี้ในรูปแบบของการตัดสดและนำไปให้กิน และการปล่อยแทะเล็ม เป็นรูปแบบที่เหมาะสมกับแพะเนื้อ โดยเพิ่มอัตราการเจริญเติบโต เปอร์เซ็นต์เนื้อแดง และมีสัดส่วน n-6 / n-3 ที่เหมาะสม (< 4)

การศึกษาครั้งนี้แสดงให้เห็นว่าพืชอาหารสัตว์เขตร้อนที่เหมาะสมต่อการเลี้ยงแพะเนื้อ คือ หญ้ามูลาโต 2 (อาหารหยาบประเภทหญ้า) และถั่วฮามาตา (อาหารหยาบประเภทถั่ว) ที่อายุการเก็บเกี่ยว 45 วันของการเจริญขึ้นใหม่หลังการตัด ในการทำพืชอาหารสัตว์หมักสารเสริม (มันเส้นบดกากน้ำตาล และ FJLB) สามารถรักษาคุณภาพพืชอาหารสัตว์ให้อยู่ในระดับดี ซึ่งการเสริมด้วย FJLB มีความสะดวกและง่ายต่อการทำพืชอาหารสัตว์หมัก และการนำไปใช้ การให้หญ้ามูลาโต 2 และถั่วฮามาตาในรูปแบบตัดสดและนำไปให้กินและในรูปแบบปล่อยแทะเล็ม เป็นรูปแบบที่เหมาะสมกับแพะเนื้อ เนื่องจากช่วยเพิ่มอัตราการเจริญเติบโต เพิ่มปริมาณเนื้อแดงมากขึ้น ทำให้เนื้อมีค่าความเป็นกรด-ด่างและความนุ่มในระดับที่ยอมรับได้ และช่วยทำให้มีอัตราส่วนของกรดไขมัน n-6 : n-3 ในกล้ามเนื้อสันนอกที่เหมาะสม

สาขาวิชาเทคโนโลยีการผลิตสัตว์ ลายมือชื่อนักศึกษา _____

ปีการศึกษา 2555 ลายมือชื่ออาจารย์ที่ปรึกษา _____

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ACHARA LUKKANANUKOOL : EFFECTS OF FORAGE SOURCES AND FEEDING REGIMES ON RUMEN FERMENTATION, NUTRIENT DIGESTIBILITY AND GOAT MEAT QUALITY. THESIS ADVISOR : ASST. PROF. PRAMOTE PAENGKOUM, Ph.D., 195 PP.

FORAGE SOURCES/FEEDING REGIMES/RUMEN FERMENTATION/MEAT QUALITY/MEAT GOATS

The aims of this thesis were to investigate suitable tropical forage sources and feeding regimes for raising meat goats. In total, 3 experiments were carried out:

Experiment I : Six tropical forages; Purple guineagrass (PG; *Panicum maximum* TD. 58), Mulato II grass (MG; *Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*), Napiergrass (NG; *Pennisetum purpureum* Schumacher), Thapra stylo (TS; *Stylosanthes guianensis* CIAT 184), Verano stylo (VS; *Stylosanthes hamata*), and Cavalcade (CC; *Centrosema pascuorum*) were harvested at 30, 45 and 60 days after regrowth. The forages were prepared as silage consisting of cassava chip, molasses and fermented juice of epiphytic lactic acid bacteria (FJLB) and untreated with additive. The results showed that dry matter yield, neutral detergent fiber and acid detergent fiber increased, but organic matter, crude protein and ether extract decreased when the cutting date was advanced. The chemical composition of the silages investigated seemed to be related to the chemical content of the fresh forage. The fatty acid (FA) profile of the forage silages was composed of key FAs as found in fresh forage, which reflected small changes in the FA pattern.

Experiment II : Eight male ruminally fistulated, Boer × Anglo-Nubian, goats were assigned in 2 × 2 factorial arrangements in 4 × 4 replicated Latin square design to receive 4 dietary treatments : Fresh MG, Fresh VS, MG silage with FJLB and VS

silage with FJLB. The results showed no effects of the dietary treatments on the rumen parameters. The main FAs of the rumen fluid were C18:0 and C18:1n9, which were derived from C18:2n6 and C18:3n3 rich in diet by ruminal biohydrogenation.

Experiment III : Thirty weaning male goats, Boer × Anglo-Nubian were used in 2 × 3 factorial arrangements in a completely randomized design with 2 forage sources (MG and VS) and 3 feeding regimes (cut-and-carry, silage with FJLB and grazing) for each forage species. The results demonstrated that the total intake of the goats raised by grazing on VS was significantly higher ($P<0.01$) than that of goats in the other groups. The meat goats raised by grazing forage had lower values of the dressing percentage and pH, while they had higher values of loin eye area and drip loss percentage. The conjugated linoleic acid (CLA) content was lowest ($P<0.05$), the n-6 : n-3 ratio was high in the loin muscle of the goats fed by silage with the forage investigated. The proportion of n-6 and n-3 FAs in the loin muscle of those groups of goats offered cut-and-carry forage and grazing was appropriate.

The overall results show that the most appropriate tropical forage source for meat goats is MG and VS, at 45 days after regrowth. All the silage additives investigated can be used to preserve the quality of forage for meat goats, however, the FJLB additive is the most practical. The cut-and-carry and grazing feeding regime for both MG and VS can be used for meat goats with a good response in many aspects, such as high growth rate, lean yield and appropriate n-6 : n-3 ratio in loin muscle.

School of Animal Production Technology
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CHAPTER I

INTRODUCTION

Diet has been recognized as a contributing factor to the development and prevention of some disease conditions (NRC, 1988). Consumers increasingly pay attention to the form and quantity of fat present in the foods they consume. This is leading to a shift in the way food is produced (Dierking et al., 2010).

The fatty acid (FA) profiles of meats have recently become increased interest due to the beneficial or detrimental impact on human health, especially unsaturated fatty acid (UFA) (Hebeisen et al., 1993). Fatty acid composition of meat has long been studied but there is still receives a lot of attention in research. The intramuscular fat composition could be partly contained with polyunsaturated fatty acid (PUFA), in particular the long chain n-3 or ω-3 (α-linolenic acid, C18:3n-3); eicosapentaenoic acid (C20:5n-3); docosapentaenoic acid (C22:5n-3) and docosahexaenoic acid (C22:6n-3) and the long chain n-6 or ω-6 FA (linoleic acid, C18:2n-6); arachidonic acid (C20:4n-6) and C22:4n-6. In general, a ratio of PUFA to saturated fatty acid (SFA) (termed P:S) above about 0.45 and a ratio of n-6 : n-3 below about 4.0 are required in human diet to combat various ‘‘lifestyle diseases’’ (Simopoulos, 2004; Williams, 2000).

Red meat is a primary dietary component and forms an important part of a balanced and varied diet (USDA, 2005). Recently, there is ample new research providing evidence that red meat can be consumed daily. However, obesity and high

SFA intake from animal products has a positive association (Biesalski, 2002; Yin and Chao, 2008). This has led to a concern that total dietary fat intake should be restricted by consuming smaller portions less frequently.

Goats meat could become an ideal choice of red meat for health conscious consumers (Carlucci et al., 1998; Johnson and Chen, 1995); due to its lower fat percentage compared to beef and lamb (Casey et al., 2003; Mahgoub et al., 2002) and a good source of PUFA (Banskalieva et al., 2000b). Goat farming is practiced worldwide, with goat products having a favorable image (Morand-Fehr et al., 2004). The number of goats has increased globally, even in countries with high and intermediate incomes (Morand-Fehr et al., 2004). In general, the intramuscular FA composition is affected by several genetic and environmental factors, amongst which the dietary supply of FA is considered to be the most important. In ruminants, a number of studies demonstrate the effect of different diets on the FA composition of total lipids in different muscles and dairy products (Dhiman et al., 1999). Therefore, quality of goat meat would be improved by dietary.

The productivity of goats in the tropical area is limited by acute shortages of good quality feed (Hove et al., 2001; Kanani et al., 2006). Poor nutrition leads to low growth performance and increased susceptibility to parasitic and other diseases. As we have known, roughage is the most important for ruminants' production. Profitable livestock production from forages depends largely on the quantity and quality of forage produced, the animal's capacity to utilize forages efficiently and ability of the livestock producers to manage forage feeding. In additional, green plants are the primary sources of C18:2n6 and C18:3n3. It is important to quantify variation in the precursors of meat and milk fat present in goat diets to identify strategies to increase

n-3, conjugated linoleic acid (CLA) or both in their productions (Chilliard et al., 2000). Thus, forage plants would represent a more natural and environmentally sustainable source of these FAs. Forages provide substantial lipids and FA in ruminant diets (Harfoot and Hazlewood, 1988). Lipids represent up to 8% of the leaf dry matter in forage plants as the report earlier (Harfoot, 1981). However, dietary PUFA are rapidly hydrogenated by the rumen bacteria, resulting in the production of SFAs (Lee et al., 2006). The predominant *trans*-11 C18:1 can be converted into *cis*-9, *trans*-11 CLA by the enzyme Δ^9-desaturase in the mammary gland and adipose tissue, and it is thought that this route forms the majority of *cis*-9, *trans*-11 CLA found in ruminant meat and milk (Lee et al., 2006; Piperova et al., 2002).

1.1 Research hypothesis

1.1.1 Six species of tropical forage**;** Purple guineagrass (*Panicum maximum* TD. 58), Mulato II grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*), Napiergrass (*Pennisetum purpureum* Schumacher), Thapra Stylo (*Stylosanthes guianensis* CIAT 184), Verano Stylo (*Stylosanthes hamata*), and Cavalcade (*Centrosema pascuorum*), harvested at 30, 45 and 60 days after regrowth, have different influences on chemical composition and fatty acid profile.

1.4.1 Silages from tropical forages harvested at 30, 45 and 60 days after regrowth with different type of additives (cassava meal, molasses and fermented juiced of epiphytic lactic acid bacteria, FJLB) were prepared to clearify the effects of feeding on fatty acid compositions of goat meat.

1.4.2 Forage sources and forms for goat meat production have different impacts on growth performance, rumen fermentation, nutrient digestibility and fatty acid composition of rumen fluid.

1.4.3 Feeding regime (grazing forage, cut-and-carry and forage silage) for meat goats have different effects on growth performance, rumen fermentation, nutrient digestibility, carcass traits, meat quality and fatty acid composition of muscle.

1.2 Research objectives

1.2.1 To investigate the nutritive values and fatty acid profiles for different species of the tropical forage harvested under different growth stages.

1.2.2 To study the effect of applying of the FJLB, in which added with cassava meal and molasses as a substrate, on the fermentative quality and chemical composition of tropical forage silages.

1.2.3 To examine the effect of forages source, silage, cut-and-carry and grazing on growth performance, rumen ecology, nutrient digestibility, fatty acid composition of rumen fluid and meat quality in goat.

1.3 Scope and limitation of this study

This study was focused on the influence of species and cutting date (days after regrowth) of tropical forages on their chemical composition and fatty acid profile, additive treatments of silages on their chemical composition and fatty acid profile and feeding regimes (grazing, silage and cut-and-carry) on rumen ecology, nutrient

digestibility and fatty acid composition of rumen fluid, tissues and meat quality in male meat goats.

1.4 Expected results

1.4.1 To optimize species and cutting date (days after regrowth) of tropical forages and improve their productivity and fatty acid composition (especially C18:2n6 and C18:3n3).

1.4.2 To optimize additive treatments of silage and improve quality of silage and their fatty acid compositions.

1.4.3 To increase the growing goats' productivity as well as meat unsaturated fatty acid content.

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CHAPTER II

LITERATURE REVIWE

2.1 Lipid in plant

Plant lipids can be grouped into structural and storage compounds. The structural lipids are present in various membranes and protective surface layers (McDonald et al., 1995). The surface lipids are mainly waxes with small proportions of long hydrocarbons, FAs and cutting. The membrane lipids, which are present in the mitochondria, endoplasmic reticulum, plastids and plasma membranes, consist mainly of glycolipids and phospholipids (McDonald et al., 1995). The synthesis of FA is thought to take place on the endroplasmic reticulum and use fatty acid esterified to the major membrane lipid phosphatidylcholine as a substrate (Figure 2.1). The five major FAs in forage are: palmitic (C16:0), stearic (C18:0), oleic (C18:1n-9), linoleic (C18:2n-6) and α-linolenic (C18:3n-3) acid, which comprise up to 95% of their total fatty acids (TFA). The FAs associated with galactolipids contain high amounts of linoleic and α-linolenic (Van Soest, 1994). The chloroplast membranes are the most abundant membranes in green leaves, comprising up to 70% of the lipids in green tissue. Thus, the galactolipids dominate the lipids in photosynthetic tissue (Taiz and Zeiger, 2002), hence are the leaves rich in α-linolenic (between 60% and 75% of TFA), linoleic and palmitic acid (6-20%), and while oleic is a minor component (Hawke, 1973).

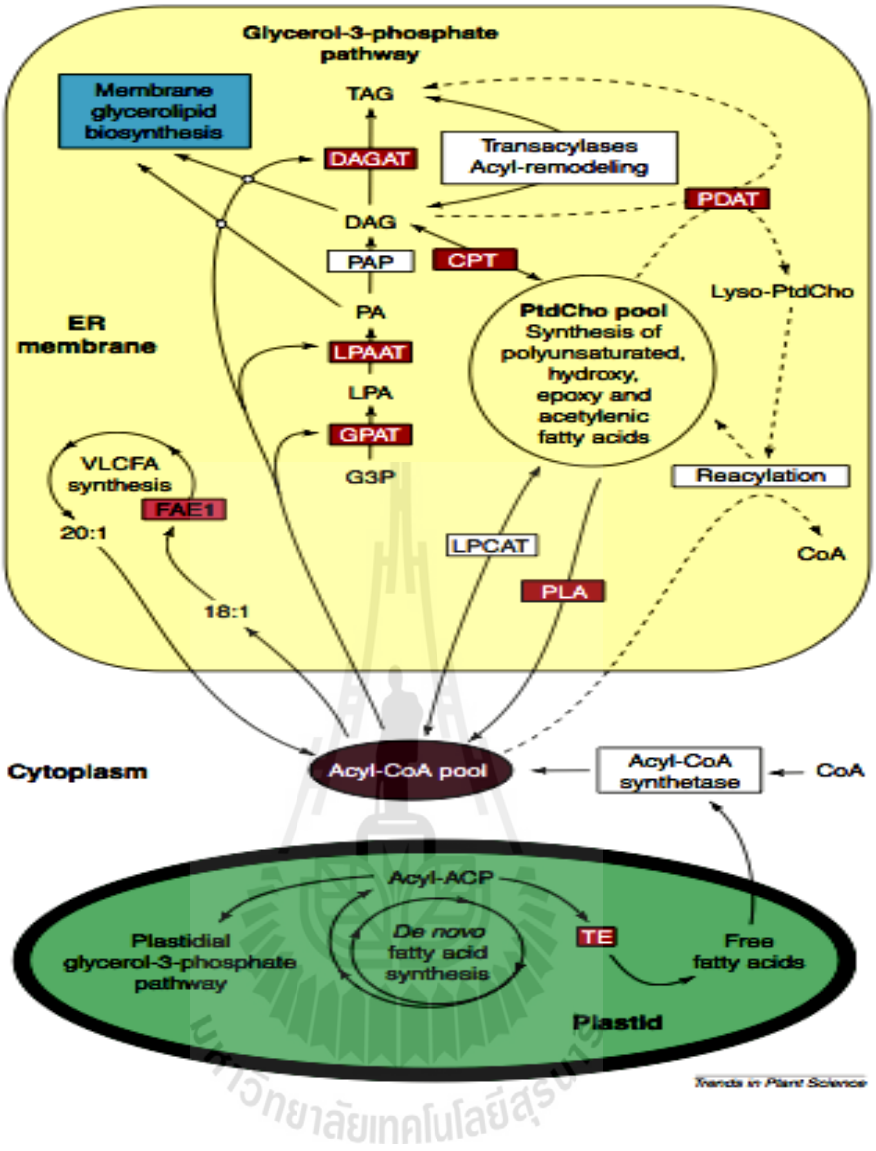

Figure 2.1 A simplified diagram of the metabolic pathways of plant lipid Biosynthesis.

Source: Millar et al. (2000).

2.2 Forage

2.2.1 Grass

In temperate countries, fresh grass contains 1-3% FA, the highest FA content being observed in spring and autumn. About 55-65% of these FA are composed of

linolenic acid (Bauchart et al., 1984). Tropical pastures have wide variation of α-linolenic acid represents 15-40% of TFA (Chilliard et al., 2001).

Jarrige et al. (1995) have reported that FA concentration in grasses increased by N fertilization. For instance, timothy is the most important perennial forage grass species in eastern Canada but little is known about the factors affecting its FA concentration. According to data presented in the companion paper (Boufaïed et al., 2003), about 51% of total FA in forages are α-linolenic acid and certain forage species may contain up to 20 mg of α-linolenic acid per gram of dry matter. Thus, managing the FA composition of grazing ruminant diets could lead to meat and milk products that have higher content of CLA, but forage FA dynamics must be more fully understood for a range of forages before grazing systems can be specified (Boufaïed et al., 2003). Considering the effects of conserved forages on beneficial milk PUFA particularly, CLA and C18:3n-3 FAs factors affecting levels of precursor PUFA in forage and forage effects on recovery of feed PUFA in milk (Dewhurst et al., 2006).

2.2.2 Legume

Legumes are a rich source of protein (both as forage and as seeds) and can be a supplementary source of nitrogen, and other nutrients, in many tropical production systems (Gutteridge and Shelton, 1994). Legumes are also useful as a nitrogen supplement in animal diets to increase basal diet efficiency. High leaf protein concentration should ideally provide a source of both fermentable and by-pass protein (Raghavan and Krishna, 1993).

Fraser et al. (2004) compared the FA profile of lambs finished on red clover, lucerne or perennial ryegrass. The data generated from the study indicated that grazing forage legumes significantly increased the proportion of linoleic and α-linolenic acid

in lamb muscle tissue and the concomitant proportion of UFAs to SFAs (0.19, 0.16 and 0.12 for lambs offered red clover, lucerne and perennial ryegrass respectively) and the n-6:n-3 ratio was 1.13, 1.08 and 0.98 for lambs grazing red clover, lucerne and perennial ryegrass, respectively.

2.3 Factors affecting fatty acid concentrations in forage

The type of dietary lipid and the dietary forage to concentrate ratio have been shown to play a crucial role in determining the products of rumen metabolism (Griinari et al., 1998) and to ultimately affect the FA profile of meat or milk fat. The n-6 : n-3 FA ration of ruminants carcasses can be influenced by FA composition of the diet fed to animals (Raes et al., 2004), moreover, the linoleic acid ratio in the diet and in the intramuscular fat were linearly relate for ruminant. Earlier studies reported that the FA composition of lipids in grasses and legumes was affected by many factors, including species and senescence (Dewhurst et al., 2001; Harfoot and Hazlewood, 1988; Harwood, 1980), and growth stage (Bauchart et al., 1984). Similar results were obtained from Dewhurst et al. (2008) who found that the effects of species, cutting date and cutting interval on the concentration of FAs in temperate grasses.

2.3.1 Species variation

Fatty acid variation within forage species is currently unknown. Previous research demonstrates that fresh forage contains high amounts of PUFA in the form of α-linolenic acid, linoleic acid and oleic acid and a sparse amount of SFAs (Hawke, 1973). While perennial ryegrass (*Lolium prenne* L.) has been most widely studied, Clapham et al. (2005) analyzed the FA content of several forage species and found that α-linolenic followed by linoleic and palmitic acid were the predominate FAs.

Dewhurst et al. (2001) compared three ryegrass species and found linoleic and α-linolenic differing by as much as 0.34 g/kg and 4.02 g/kg, respectively. Similarly, Boufaïed et al. (2003) found legumes contained 1.3 times more linoleic acid than grass species while grasses contained 1.1 times more α-linolenic acid than legumes on average.

2.3.2 Stage of maturity

The most important factors influencing the FA content of fresh forage appear to be cutting date and interval, which reflect maturity differences. The advances in the stage of maturity, there were a trend of lower SFA and MUFA, essentially in agreement with Vanhatalo et al. (2007), SFA and PUFA concentration in timothygrass (*Phleum pratense*) and red clover (*Trifolium protense*) were higher in early cut than late cut. Also Clapham et al. (2005) found that in grasses (orchardgrass and perennial ryegrass) and legume (white clover) decreased the concentration of SFA, MUFA and PUFA with plant maturity, confirming observation from Vanhatalo et al. (2007).

2.3.3 Preservation

Ensiling is one of the best ways to preserve green forage by controlling anaerobic fermentation (Islam et al., 2001; Bureenok et al., 2005; Khan et al., 2009) The success of the ensiling can be achieved when the number of lactic acid bacteria (LAB) is dominant in the fermentation. Lactic acid bacteria utilizes water-soluble carbohydrate (WSC) to produce lactic acid (LA), the primary acid, responsible for decreasing the pH in silage. Quick reductions in silage pH will inhibit the growth of undesirable anaerobic microorganisms such as enterobacteria and clostidia (Bureenok et al., 2005; Driehuis et al., 2000; Kung, 2000). However, ensiling reduces the positive effects of herbage lipids on the FA composition of ruminants' production due to

extensive hydrolysis of forage lipid in the silo (Dewhurst et al., 2006; Chilliard et al., 2007; Khan et al., 2009).

Plant lipases release free fatty acid (FFA) from damaged tissues after cutting (Thomas, 1986), or during ensiling, of herbage (Chow et al., 2004). Free fatty acid can be further oxidized by plant lipoxygenases (Fall et al., 1999; Feussner and Wasternack, 2002). Some studies report a decrease on the total FA content of ryegrass silages compared with those of fresh products (Dewhurst and King, 1998; Elgersma et al., 2003), probably when undesirable fermentations occur (Lough and Anderson, 1973) or when silage is wilted (Dewhurst and King, 1998). However, if herbage is ensiled directly after cutting (Ueda et al., 2002), or wilted for only a short time (<24 h), the concentration of FA remain relatively stable (Arvidsson et al., 2009).

2.3.3.1 Additive treatments

The tropical forage is difficult primarily due to deficiency of WSC (McDonald et al., 1991). Silage additives can be used when ensiling problem or 'at risk' forages to improve silage fermentation quality, reduce ensiling losses and improve silage nutritive value. However, inoculants have been shown to improve animal production, even where silage is well preserved without an additive (Van Ranst et al., 2009; Alves et al., 2011). Van Ranst et al. (2009) reported that in the silages was no effect of additive on total FA content.

Table 2.1 Concentration of fatty acids in plants (% of total FAs).

Plant	Harvest*	Lauric	Myristic	Palmitic	Palmitoleic	Stearic	Oleic	Linoleic	α-linolenic	Total	Reference
Grass	-	-	1.35	6.04	0.69	0.95	1.66	4.06	14.25	29.0	French et al. (2000)
Grass silage	-	-	1.52	6.72	0.16	0.81	1.7	4.07	12.94	28.0	
Triticale	1	0.022	0.55	5.43	1.16	0.25	0.94	5.17	30.0	43.5	Clapham et al. (2005)
	2	0.064	0.38	3.83	0.56	0.19	0.62	3.38	19.4	28.4	
	3	0.066	0.26	3.04	0.35	0.14	0.52	2.73	13.2	20.3	
Orchard grass	1	0.031	0.56	6.81	1.19	0.30	1.10	7.97	34.4	52.3	
	2	0.043	0.50	5.49	0.80	0.27	0.65	5.84	27.1	40.7	
	3	0.077	0.41	4.41	0.56	0.23	0.42	4.66	21.0	31.7	
Perennial ryegrass	1	0.027	0.62	6.99	0.94	0.30	1.46	6.76	34.7	51.8	
	2	0.046	0.62	6.30	0.74	0.32	1.01	5.74	31.5	46.3	
	3	0.072	0.61	5.91	0.56	0.28	0.71	5.47	26.8	40.5	
White clover	1	0.019	0.42	6.52	1.01	0.54	1.40	8.23	26.7	44.8	
	2	0.023	0.42	5.62	0.75	0.47	0.89	5.89	20.3	34.4	
	3	0.104	0.51	4.85	0.59	0.44	1.21	6.27	17.8	31.8	

* Harvest 1, 2, 3

Table 2.1 (Cont.) Concentration of fatty acids in plants (% of total FAs).

Plant	Harvest*	Lauric	Myristic	Palmitic	Palmitoleic	Stearic	Oleic	Linoleic	α-linolenic	Total	Reference
Chicory (Forage Feast)	1	0.014	0.46	7.63	1.33	0.29	1.33	10.69	39.6	61.3	
	2	0.016	0.45	6.32	0.88	0.25	0.48	8.08	25.6	42.1	
	3	0.007	0.42	5.69	0.78	0.25	0.33	6.42	25.0	38.9	
Chicory (Lacerta)	1	0.019	0.43	7.16	1.28	0.29	1.18	10.49	35.3	56.2	
	2	0.024	0.38	5.42	0.74	0.25	0.40	6.88	21.1	35.2	Clapham et al. (2005)
	3	0.024	0.27	4.64	0.51	0.24	0.48	5.74	14.8	26.7	
Chicory (Puna)	1	0.013	0.46	7.39	1.25	0.25	1.24	9.69	42.5	62.8	
	2	0.030	0.42	5.65	0.81	0.22	0.43	7.17	24.2	38.9	
	3	0.013	0.35	5.01	0.63	0.22	0.31	5.88	19.8	32.2	
Grass	-	-	-	4.27	-	1.26	0.67	3.35	14.25	30	Noci et al. (2007)
Grass silage	-	0.05	0.12	2.78	-	0.31	0.05	2.19	5.52	12.3	Vlaeminck et al. (2006)
Grass hay	-	-	0.27	1.78	0.19	0.24	1.32	2.56	3.82	9.98	Demirel et al. (2006)

* Harvest 1, 2, 3

2.4 Biohydrogenation in ruminant

2.4.1 In rumen

The rumen ecology of ruminants is dependent on the fermentation of its feed constituents by the rumen microorganisms (Williams and Orpin, 1987). Other mechanisms affecting ruminal biohydrogenation appear to operate through effects on lipolysis process. Lipolysis of ester linkages of dietary lipids is the initial step in lipid metabolism in the rumen. In the rumen, dietary lipids are subject to hydrolysis by microbial lipases followed by biohydrogenation of the unsaturated FFA by rumen bacteria. In the case of linoleic acid, the end product of the hydrogenation is stearic acid (Jenkins, 1993). When biohydrogenation does not go to completion, intermediates (e.g., *trans*- C18:1) from the incomplete biohydrogenation of PUFA become available for deposition in microbial biomass as well as in animal tissues (Bauman and Griinari, 2003; Or-Rashid et al., 2009) (Figure 2.2). One of the simplest approaches to reduce rumen biohydrogenation is altering the rumen microflora by reducing rumen pH. This effect has been achieved in a number of studies with diets containing a high proportion of starch-rich concentrates (Kalscheur et al., 1997; Kucuk et al., 2001; Piperova et al., 2002). Generally this militates against increasing milk PUFA in high-forage systems since forages usually lead to a relatively high rumen pH. In this case, the mechanism appears to be selective inhibition of several strains of *Butyrivibrio fibrisolvens* (Min et al., 2005), one of the most important biohydrogenating ruminal bacteria species.

In ruminants, n-6 and n-3 FA containing in dietary fats are hydrolyzed by rumen microorganisms and hydrogenated to mainly stearic acid. Small amounts taken up by the microbes will escape hydrogenation in the fore-stomachs, and absorbed and deposited in the tissues or transferred to meat and milk (Jakobsen, 1999). Despite the

primary product of biohydrogenation of C18:2n-6 and C18:3n-3 also yields a variety of MUFA, dienoic, or trienoic FA intermediates with *cis* or *trans* double bonds. After isomerization of C18:2n-6 to *cis9, trans-11* C18:2, sequential reductions of double bonds at carbons 9 and 11 yield *trans11*-C18:1 and C18:0.

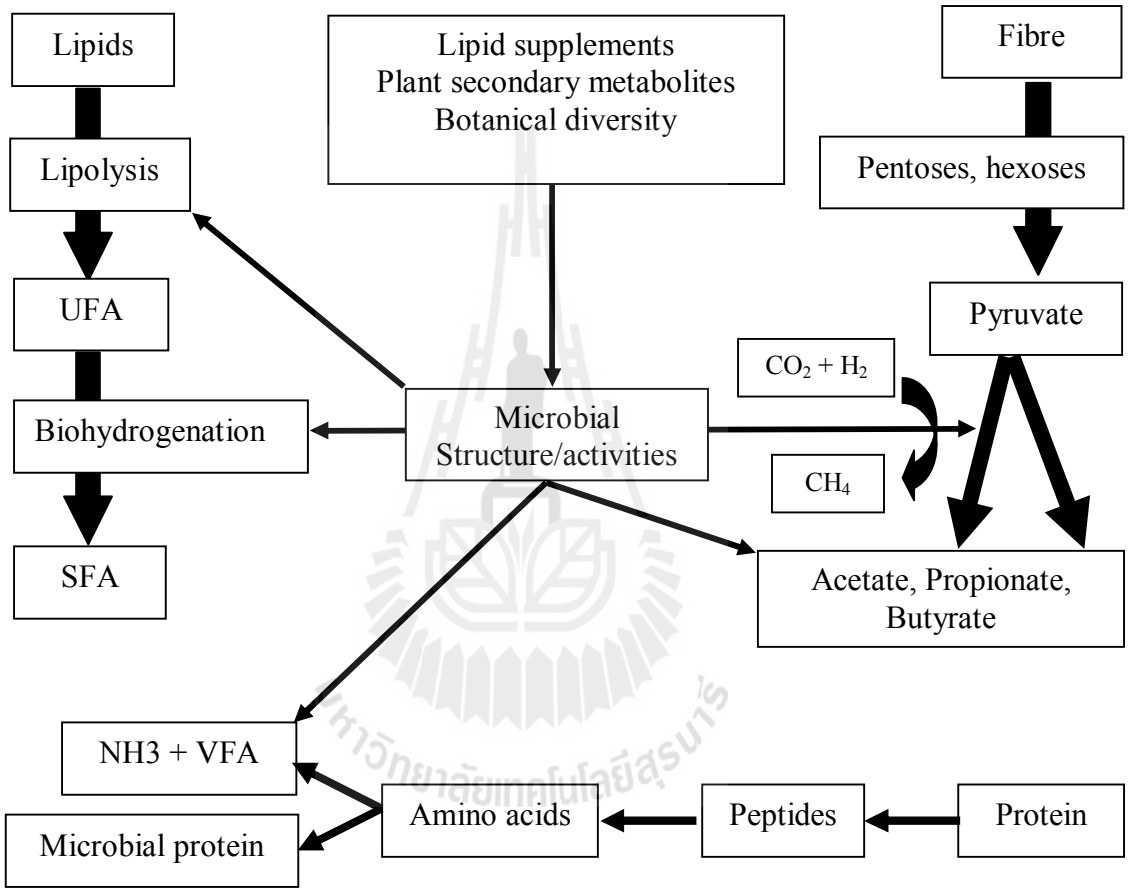

Figure 2.2 Interventions to manipulate lipid metabolism in the rumen inevitably lead to effects on other processes. Sometimes the target organisms have several functions, in other cases the metabolic pathways are linked, for example by the availability of H2. UFA: unsaturated fatty acid; SF: saturated fatty acid; VFA: volatile FA.

Source: Lourenço et al. (2010)

Biohydrogenation of C18:3n-3 also requires an initial isomerization to form a conjugated triene, followed by sequential reductions of double bonds at carbons 9, 15, and 11 to yield *trans-11, cis-15* C18:2, *trans-11* C18:1, and C18:0 (Loor and Herbein, 2003). Amounts of biohydrogenation intermediates produced in the rumen influence their concentrations in tissues or milk (Loor et al., 2002). Concentrations of *trans-11* C18:1 and *cis- 9, trans-11* C18:2, for example, are greater in milk or meat (Shen et al., 2007) from grazing cattle (Banskalieva et al., 2000a; Loor et al., 2002; Piperova et al., 2002). In a review of Wood et al., 2008, they reported that a variable proportion of dietary C18:3n-3 is biohydrogenated about 85-100% but this is more than for C18:2n-6 about 70-95%, so less is available for incorporation into tissues. Lipolysis *in vitro* has been reported to be decreased by advances in forage maturity (Gerson et al., 1968), and there is evidence that drying, versus ensiling, also reduces the extent of hydrolysis of forage lipids (Boufaïed et al., 2003).

2.4.2 In muscle and adipose tissue

The FA composition of adipose tissues and muscles of young goats was shown to reflect the FA composition of their milk intake (Bas et al., 1987; Sauvant et al., 1979), while the tissue FA composition of older goats results from changes in the activity of rumen bacteria with an increase in total saturated FA contents and the presence of odd chain-length FA, branched-chain FA, trans FA and CLA isomers. Some studies have shown that goats and sheep, fed cereal-based diets, produced abnormally high contents of odd-chain FA and methyl branched chain FA in subcutaneous adipose tissues (Bas et al., 1980; Duncan et al., 1976). The FA composition of rumen bacteria is characterized by a large proportion of SFA (Vlaeminck et al., 2006). In addition, FA of rumen bacteria contains various MUFAs

such as *trans-10* C18:1 & *trans-11* C18:1 and C18 PUFA such as *cis-9, trans-11* C18:2, *trans-10, cis-12* C18:2 & *trans-15* C18:2 (Kucuk et al., 2001; Loor et al., 2005) derived from hydrogenation of dietary C18:2n-6 and C18:3n-3.

2.5 Fatty acid composition in meat and adipose tissue

It is well established that the FA composition of muscle lipids has an important impact on meat flavour, because lipid degradation can produce aldehydes, which influence the flavour of meat at cooking. However, in studies ruminant nutritionist show that different nutritional condition can change muscle lipid FA composition, PUFA level and the n- 3:n-6 PUFA ratio (Banskalieva et al., 2000a). The UFAs that escape from the rumen are absorbed from the intestine into the circulation system, where they are transported to the mammary gland and adipose tissue and then used in the synthesis of triacylglycerols and phospholipids (Griinari and Bauman, 1999). Ruminants' mammary glands and adipose tissues contain the enzyme Δ^9-desaturase (Bauman et al., 2000; Griinari and Bauman, 1999), which introduces a *cis*-double bond between carbons 9 and 10 in FAs. The adipose tissue seems to be the major site of endogenous synthesis of CLA in growing animals, but in lactating ruminants the mammary gland is the main apparent site of endogenous synthesis of CLA (Kinsella, 1972; Bickerstaffe and Annison, 1970). Conjugated linoleic acid has health benefits in the human diet although meat from ruminants makes only a small contribution towards nutritionally significant levels.

Fat content and FA composition are important aspects of nutritional quality. A low level of fat is desirable; the balances of FAs are also important. Although sheep meat is relatively high in SFA because of hydrogenation of dietary fat in the rumen, it

has significant concentrations of linolenic acid and other n-3 PUFA including eicosapentaenoic acid (C20:5n-3, EPA) and docosahexaenoic acid (C22:6n-3, DHA) (Enser et al., 1998). The ratio between n-6 PUFA (formed from linoleic acid, C18:2n-6) and n-3 PUFA is an important nutritional index and this is within the recommended range (<4.0) in sheep meat (Enser et al., 1998). Sheep meat is a good source of CLA, which has anti-carcinogenic properties and other benefit for human health, mainly the *cis-9, trans-11* isomer (Wachira et al., 2002).

Table 2.2 Fatty acid composition (%) of total lipids in different goat muscles (mean% of pooled data).

Fatty acid: muscle/species	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	C20:4	SFA	MUFA	PUFA	Goat breed	Age (week)
Brachii (Sauvant et al., 1979)	1.20	15.41	0.39	14.49	41.66	13.67	-	-	38.76	43.51	13.67	A (Matsuoka et al., 1997)	5±22
Leg (Nitsan et al., 1987)	4.85	15.60	7.27	14.95	28.00	11.50	-	2.05	29.19	57.79	13.10	S (Johnson et al., 1995)	5±10
Rib-LD (Potchoiba et al., 1990)	5.05	31.35	5.65	14.95	28.00	11.50	1.20	-	53.80	33.65	12.70	A (Nitsan et al., 1987)	20
LD (Park and Washington, 1993)	2.93	22.30	4.73	16.20	46.20	9.23	-	3.43	41.43	50.93	12.66	A	20
LD (Park and Washington, 1993)	3.58	23.10	2.40	17.20	36.20	11.80	-	4.67	43.88	42.30	16.47	N	20
BF (Park and Washington, 1993)	2.56	21.40	1.30	15.90	39.30	15.10	-	4.52	39.86	42.00	19.62	A	20
BF (Park and Washington, 1993)	4.76	24.00	4.50	13.90	38.70	8.06	2.18	3.54	44.01	46.78	13.78	N	20
Leg (Johnson and Chen, 1995)	2.13	26.50	4.00	16.77	39.80	4.27	1.43	2.00	48.50	43.80	7.80	F (Potchoiba et al., 1990)	24±32
LT (Matsuoka et al., 1997)	1.97	20.65	3.00	11.79	47.86	7.44	0.71	2.15	35.54	53.04	11.27	JS (Potchoiba et al., 1990)	36±40

Goat breeds - A: Alpine; F: Florida; N: Nubian; S: Saanen; JS: Japanese Saanen. Muscles - BF: biceps femoris; LD: longissimus dorsi; LT: longissimus thoracis; SM: semimembranosus; TB: triceps brachii; GM: gluteus medius; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids. **Source:** Modified from Banskalieva et al. (2000).

2.6 Feeding and utilization

2.6.1 Fresh forage

Forages provide a low cost approach in comparison with diet supplementation strategies, such as oils and starch, which are designed to improve milk or meat FA profiles (Dewhurst et al., 2003). Moreover, the highly USFA are major constituents of the saponnifiable fraction of lipids extracted from green plants. The presence of PUFA in alfalfa leaf meal and buckwheat leaf meal has been established. As linoleic and α-linolenic acids are the principal PUFA in the green plants and meals, the consumption of such materials would provide the FA considered being essential in the nutrition of farm animals (Ward et al., 2002).

2.6.1.1 Cut-and-carry

Cut-and-carry feeding is labor-intensive, engaging farmers for up to six hours each day. Forage is therefore the most expensive input to animal production. Despite this, farmers offer high levels of forage to their animals, allowing them to reject around 40%. This strategy is termed ‘excess feeding’ and improves the quality of the diet consumed. Cut-and-carry forage system was used in small livestock in tropical Asia, based on nitrogen-fixing forage crops. Nitrogen-fixing farming systems are those which integrate nitrogen-fixing plants into as many parts of the system as possible (Palmer, 1996). The hypothesis is that the effective and efficient utilization of nitrogen-fixing trees and/or shrubs, as well as other nitrogen-fixing plants, can contribute to the overall sustainability of farming systems because of the addition of extra N through biological fixation.

However, cut-and-carry systems extract a considerable amount of nutrients from the forage production area and these are moved to where the animals are fed; particular care is required to return nutrients to the forage area.

2.6.1.2 Pasture feeding

Effects of forage species and cultivar described above for conserved forages are also relevant to grazed pasture. Indeed, genetic differences in the FA concentrations will be more apparent in young growing plants, versus when flowering and senescence become important in more mature grasses destined for conservation as silage or hay. (Nudda et al., 2005) reported that the FA profiles of milk and dairy products from sheep grazing pasture had lower concentrations of C18:3 n-3, *cis-9, trans-11* CLA and *trans-11* C18:1 during spring and summer. The authors attributed this effect to declining quality and quantity of pasture. Moreover, access to pasture for just 8 hours led to significant increases in concentrations of C18:3 n-3, *cis-9, trans-11* CLA and *trans-11* C18:1 for cows offered a total mixed ration (Loor and Herbein, 2003).

Dannenberger et al. (2004) examined the effect of grazing bulls on grass pasture in comparison to the intensive feeding of indoor bulls with concentrate, in order to enhance the concentration of n-3 FAs in bulls muscle. Pasture-fed lambs had a significantly higher proportion of CLA in the muscle and total n-3 in muscle lipids and a reduced ratio of n-6:n-3, compared to concentrate-fed lambs. Consistent with these findings, comparing the FA profile of the muscle of grass-fed lambs with that of concentrate and hay-fed lambs, (Aurousseau et al., 2004) found that grass-fed lambs produced meat with a higher CLA content and greater ratio of n-3:n-6. French et al., 2000 determined in the intramuscular fat of steers (*longissimus dorsi* muscle)

increasing CLA contents consistent with increasing intakes of grass. Additionally, grass silage also positively influenced CLA content but not to the same extent. (Poulson et al., 2004) reported a higher CLA content in the *longissimus* and *semitendinosus* muscle from steers raised only on forages compared to steers fed a common high grain feedlot. Steers fed a grain based diet in the growing period and grazed on pasture during the finishing period still had a higher CLA tissue content compared to those fed only the grain based diet.

In addition, grazing on pasture for about 200 days and then being shifted to a dry lot diet for about 60 days also led to significantly higher CLA concentrations in steers and heifers compared with animals offered only the dry lot diet (Sonon et al., 2004). Contrary to these results of Dannenberger et al. (2005) who found no significant effect of grass feeding on the CLA content in bulls and steers compared with concentrate feeding. However, in a subsequent study (Nuernberg et al., 2004) reported significantly higher proportions of the *cis9, tran11* C18:2 isomer in bulls and lambs after pasture feeding compared with concentrate feeding. In agreement with Santos-Silva et al. (2002) who reported higher CLA concentrations in the *longissimus dorsi* muscle of lambs raised on pasture than that of lambs fed a concentrate diet. Aurousseau et al. (2004) noted that CLA content in muscle triglycerides depended not only on the diet but also on the growth rate. Again CLA concentration was higher in grass fed lambs compared to those fed the concentrate and was even higher at higher growth rates. This may be due to the higher daily grass intakes of these lambs.

Pasture feeding does not only cause higher CLA concentrations but also influences FA composition. A decrease in the n-6:n-3 PUFA ratio as well as an increase in the PUFA:SFA is described in beef adipose and muscle tissue by inclusion

of grass in the diet (French et al., 2000; Realini et al., 2005). In lambs, a decrease in n-6:n-3 PUFA ratio has been documented as well as increase in the PUFA:SFA is described in beef adipose and muscle tissue by inclusion of grass in the diet (Aurousseau et al., 2004; Santos et al., 2008). The increased CLA content in meat from animals grazing on pasture is attributed to the high PUFA content of grass (especially C18:3n3 with an n-6:n-3 ratio of approximately 1:3-5). Although the amount of dietary PUFA determines the generation of *trans* FAs by rumen bacteria as discussed earlier (Lawson et al., 2001). However, this alone does not explain why hay and grass silage differ in the magnitude of CLA production.

2.6.2 Silage forage

Goats are natural browsers in the wild, being very selective of what they eat. If the seasonal nutritive values of browse and other feedstuffs decline or fluctuate, silage can be a good alternative source for goats (Hibma, 2008). The dominant FA composition of perennial ryegrass silage was C16:0 and C18:3n-3 and considerably lower concentration of oleic acid (*cis-9* C18:1) and linoleic acid (C18:2n-6). Conversely, the silage was high in α-linolenic acid (C18:3n-3) (Warren, 2008).

Increasing use of silage made from grass and legume emphasized the need for data on the composition of this type of livestock feed. The patiently of information concerning the levels of PUFA remaining in the silage after the fermentation has take place led to the present investigation. Van Ranst et al. (2009) have reviewed that the majority of the FA in silages occurs as FFA due to lipolysis process. A range of studies have shown a higher n-3 FA content of milk and meat of ruminants fed clover silages in stead of pure ryegrass silages. The higher n-3 FA content in products of clover silage fed ruminants cannot always be appointed to a higher dietary supply.

Red clover feeding was associated with a higher post ruminal recovery of n-3 FA (Lee, 2003) and a lower rumen biohydrogenation (Loor and Herbein, 2003).

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CHAPTER III

EXPERIMENT I

3.1 Eexperiment I-I : Comparative study of different species and cutting date on the chemical composition and fatty acid profile tropical forage

3.1.1 Abstract

This experiment was carried out to investigate dry matter (DM) yield, chemical composition and fatty acid (FA) composition for different species of the tropical forages; Purple guineagrass (*Panicum maximum* TD. 58), Mulato II grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*), Napiergrass (*Pennisetum purpureum* Schumacher), Thapra Stylo (*Stylosanthes guianensis* CIAT 184), Verano Stylo (*Stylosanthes hamata*), and Cavalcade (*Centrosema pascuorum*) harvested at 30, 45 and 60 days after regrowth. The studied tropical grasses were detected significant effect of cutting date for higher DM yield ($P<0.01$), higher DM ($P<0.01$), lower OM ($P<0.01$), lower EE ($P<0.01$), higher NDF ($P<0.001$) and higher ash ($P<0.001$) with advancing regrowth age, without significant difference for species and interaction of species and cutting date. For legume species, there were significant effects of legume species for the DM yield ($P<0.001$) and the content of DM ($P<0.001$), OM ($P<0.05$) and ash ($P<0.001$). As regrowth age of all three legumes increased, there were mainly higher DM yield ($P<0.001$), NDF ($P<0.01$), ADF ($P<0.01$) and ash ($P<0.001$) contents

while the contents of OM, CP and EE were decreased (P<0.001) with increase of cutting date. The main FA compositions were C16:0 (palmitic acid), C18:0 (stearic acid), C18:1n9 (oleic acid), C18:2n6 (linoleic acid) and C18:3n3 (α-linolenic acid). Grass species factor had significant difference (P<0.001) for C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3. For legume forage, the proportion of C16:0 and C18:1n9 had no influence of legume species, whilst there were significant difference (P<0.001) for the proportion of C18:0, C18:2n6 and C18:3n3. The factor of cutting date (days after regrowth) was found highly significant difference (P<0.01) for the proportion of C16:0 and C18:0 and very highly significant difference (P<0.001) for the contents of C18:1n9, C18:2n6 and C18:3n3. The experimental grasses and legumes would increase DM yield and content of DM, NDF, ADF and ash, but decrease OM, CP and EE with advancing maturity of the forage. However, there are inconsistent pattern of FA profile in forage. The Mulato II grass and Verano stylo with the harvesting at 45 days after regrowth would contain proper chemical composition and properties including proper FA profile for ruminants.

Key Words: Tropical grasses and legumes, Regrowth age, Dry matter yield, Chemical composition, FA composition

3.1.2 Introduction

Roughage is the most important for ruminants' production. Profitable production of ruminants depends largely on the quantity and quality of obtained forage. In addition, forage plants represent a more natural and environmentally sustainable source of FAs for ruminants (Harfoot, 1981; Mel'uchovà et al., 2008).

Forages provide a low cost approach in comparison with diet supplementation strategies, such as oils, weed grain, seed and starch etc., which are designed to improve animal production (milk or meat) and FA profiles (Dewhurst et al., 2003).

Even though forages contain relatively low amounts of lipids but there are important source of precursor of PUFA and CLA in milk and meat of ruminants, such as linoleic acid and α- linolenic acid (Vanhatalo et al., 2007; Dewhurst et al., 2006; Wyss et al., 2006; Harfoot and Hazlwood et al., 1988). The C18:2n6 and C18:3n3 are the principal PUFA in the green plants, the consumption of such materials would provide the FAs considered being essential in the nutrition of farm animals (Ward et al., 2002). Although it has been established that forage-base diets alter the FA composition of meat and milk products but a few studies have investigated the relation between the forages species and FA composition (Dierking et al., 2010 and Morand-Fehr et al., 2007).

Previously studies reported that the FA composition of lipids in grasses and legumes was affected by many factors, including species and senility (Harfoot and Hazlewood, 1988; Harwood, 1980), growth stage (Bauchart et al., 1984), conservation method (Yang and Fujita, 1997; Lough et al., 1994), as well as wilting, shading, and silage additives (Dewhurst and King, 1998). Dewhurst et al. (2001) reported that plant species, cutting date, and cutting interval have a significant impact on PUFA concentrations in forage. Similar results were obtained from Dewhurst et al. (2008) who found the effects of species, cutting date and cutting interval on the concentration of FAs in temperate grasses. These factors would also affect to FA composition of forage grown in tropical area. There is a little information about FA profile of the forage used in farms in Thailand.

The present study, therefore, was conducted to examine the effect of species and cutting date (days after regrowth) on chemical composition and quantify the variation in FA content of the tropical forage.

3.1.3 Material and methods

3.1.3.1 Plant materials

The grasses and legumes evaluated in this study as the followings: three grass species; Purple guineagrass (*Panicum maximum* TD. 58), Mulato II grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*) and Napiergrass (*Pennisetum purpureum* Schumacher) and three legume species; Verano Stylo (*Stylosanthes hamata*), Thapra Stylo (*Stylosanthes guianensis* CIAT 184) and Cavalcade (*Centrosema pascuorum*). A series of 30 plots (each 3 m × 3 m) was sown without fertilizer on February 2008 at Faculty of Natural Resources, Rajamangala University of Technology - Isan, Sakon Nakhon Campus according to a 3 × 3 factorial arrangement in completely randomized design (CRD). Samples were taken in May, July and September 2008 at 30, 45 and 60 days after regrowth. After cutting in each date, fertilizer was not applied.

3.1.3.2 Chemical analysis

For forage quality at harvesting, the forage in a measured area of 1 m^2 was hand clipped and weighed. Each subsample was dried in a hot-air oven at 60 °C to determine dry matter (DM) content, then ground to pass through a 1 mm^2 mesh screen and analyzed for chemical composition. Total N was determined using the Kjeldahl method and crude protein (CP) was calculated by multiplying the N content by 6.25, Ether extract (EE) and ash contents were quantified by AOAC (1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) estimated by the methods

described by Goering and van Soest (1970).

3.1.3.3 Fatty acid methyl ester of oil sample

After cutting, the fresh forage samples were immediately frozen at -20 °C and subsequently freeze-dried and prepared for FA analysis by gas chromatography (GC) of fatty acid methyl ester (FAME). The lipids were extracted from the forages using the chloroform/methanol (70:30) method procedure of Folch et al. (1957) and Methylation of oil samples was used as by described by the procedure of Metcalfe (1966). Fatty acid composition was measured after methylation of samples. FAME were analyzed on a Perkin Elmer Auto system GC equipped with a flame-ionization detector (FID) using a capillary column (SPTM - 2560, 100 m × 0.25 mm ID, 0.20μm film). This analyzed adopted a split injection (split ratio 100:1). The GC analysis was temperature programmed, at 140 °C held at 5 min, and raised from 140 °C to 240 °C at a rate of 4 °C/min and then held 240 °C for 40 min. The injection port and detector temperatures were set at 260 °C. Helium was used as the carrier gas at a rate of 20 cm/sec. Identification of the FA was based upon retention times using standards of methyl esters. A mixture of the standards of the individual FAME was used to determine response factors. The areas of the peaks in the chromatogram were calculated and normalized using response factors. Proportions of individual FA were calculated.

3.1.4 Statistical analysis

Data were statistically analyzed according to 3 × 3 factorial arrangements in completely randomized design of each forage using the PROC GLM procedure (SAS, 1990). Significant differences ($P<0.05$) among treatments were determined using

Duncan's News Multiple Range test according to Steel and Torrie (1980).

3.1.5 Results

3.1.5.1 Dry matter yield and Chemical composition of forage

The DM yield and chemical compositions of the experimental grasses are shown in Table 3.1.1 For the concentration of chemical composition, the studied tropical grasses were detected significant effect of cutting date for higher DM yield ($P<0.01$), higher DM ($P<0.01$), lower organic matter (OM) ($P<0.01$), lower EE ($P<0.01$), higher NDF ($P<0.001$) and higher ash ($P<0.001$) when the regrowth age increased, without significant difference for species and interaction of species and cutting date. This also found influence of days of regrowth for the higher ADF ($P<0.001$) with higher cutting date, but the interaction was presented ($P<0.05$). For the CP contents, there was highest CP content (7.26%) for the Mulato II grass at 45 days of regrowth ($P<0.01$) compared with the Purple guineagrass and Napiergrass at 45 days of regrowth (5.62% for both). The contents of CP decreased ($P<0.001$) with higher days after regrowth, except for the Multo II at 45 days of regrowth still containing high CP content. However there was no influence of the interaction for CP contents. On overview, all three grasses had higher DM yield and proportion of DM, NDF, ADF and ash, but lower for content of OM, CP and EE, when the regrowth age increased.

For the DM yield and chemical composition of the studied legumes species, as shown in Table 3.1.2. When legume species factor as considered, there were significant effect of legume species for the DM yield ($P<0.001$) and the content of DM ($P<0.001$), OM ($P<0.05$) and ash ($P<0.001$). The DM yield of the Cavalcade was the

lowest when compared to Thapra stylo and Verano stylo. The DM content in Verano stylo and Cavalcade had higher content of DM at 60 and 45 days after regrowth, respectively. The Verano stylo had the lowest CP content at 60 days of regrowth, while the trend of lower CP content was found in the Cavalcade at 45 days after regrowth. The content of ash was highest in the Verano stylo at 60 days after regrowth. The regrowth age of all three legumes increased, there were mainly higher DM yield ($P<0.001$), NDF ($P<0.01$), ADF ($P<0.01$) and ash ($P<0.001$) contents while the contents of OM, CP and EE were decreased ($P<0.001$) with increase of cutting date. The interaction between legume species and cutting date was found ($P<0.05$) for the ADF content. On overview, all three legumes had rather similar pattern of DM yield and chemical composition to those of the three grasses. They also had high DM yield and content of DM, NDF, ADF and ash, but low for content of OM, CP and EE, when the regrowth age increased.

3.1.5.2 Fatty acid composition of forage

The FA composition (g/100 g total fat) of the tropical grasses and legumes has demonstrated in Table 3.1.3 and 3.1.4, which the main FA compositions were C16:0 (palmitic acid), C18:0 (stearic acid), C18:1n9 (oleic acid), C18:2n6 (linoleic acid) and C18:3n3 (α-linolenic acid).

Grass species factor had significant difference ($P<0.001$) for C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3. There was inconsistent proportion for the C16:0 and C18:3n3 among the grasses. The content of C18:0 was higher in the Purple guineagrass compared with the Mulato II grass and Napiergrass. The Mulato II grass contained higher C18:1n9 than the other two grasses, except for the Mulato II at 30 days of regrowth. The C18:2n6 content in the Mulato II grass was higher than that

in the Purple guineagrass and the Napiergrass. For the cutting date effect, there were decreased proportions ($P<0.001$) of C16:0 and C18:3n3 when the cutting date increased, whereas the contents of C18:0 ($P<0.05$), C18:1n9 ($P<0.001$) and C18:2n6 ($P<0.01$) were no systematic pattern with higher cutting date. The interaction between grass species and cutting date was found for C18:1n9 and C18:2n6. For grouped FAs, there were no systemic patterns for SFA, MUFA and PUFA for species, cutting date and their interactions, except for high MUFA in Mulato II grass at 45 and 60 days after regrowth. The ratio of PUFA/SFA was also no systemic pattern with all factor effects, but all ratios were more than 1 (1.69 - 2.51).

Table 3.1.1 Effect of species (S) and cutting date (D, days of regrowth) on DM yield (ton/ha) and chemical composition (% on DM basis) in the tropical grasses.

Item	Purple guineagrass			Mulato II grass			Napiergrass			SEM	S	D	S × D
	30 D	45 D	60 D	30 D	45 D	60 D	30 D	45 D	60 D				
DM yield	3.39 ^{CD}	5.59 ^{ABCD}	6.81 ^{ABCD}	4.29 ^{BCD}	8.28 ^{ABC}	9.46 ^{AB}	2.79 ^{D}	8.02 ^{ABC}	10.06 ^{A}	0.478	ns	**	ns
DM	25.55 ^{AB}	29.42 ^{A}	30.26 ^{A}	22.96 ^{AB}	29.12 ^{A}	28.75 ^{A}	20.35 ^{B}	28.94 ^{A}	31.14 ^{A}	0.779	ns	**	ns
OM	86.48 ^{A}	85.69 ^{AB}	79.68 ^{CD}	85.57 ^{AB}	84.28 ^{ABC}	75.27 ^{D}	87.61 ^{A}	80.09 ^{BCD}	75.41 ^{D}	0.501	ns	***	ns
CP	7.27 ^{A}	5.62 ^{B}	4.07 ^{CD}	7.10 ^{A}	7.26 ^{A}	4.39 ^{C}	7.13 ^{A}	5.62 ^{B}	3.18 ^{D}	0.123	**	***	ns
EE	2.18 ^{A}	1.78 ^{AB}	1.25 ^{B}	2.20 ^{A}	1.85 ^{AB}	1.61 ^{AB}	2.49 ^{A}	1.86 ^{AB}	1.60 ^{AB}	0.081	ns	**	ns
NDF	55.75 ^{D}	71.90 ^{ABC}	84.12 ^{A}	54.89 ^{D}	65.44 ^{BCD}	76.90 ^{AB}	59.65 ^{CD}	61.22 ^{CD}	81.22 ^{A}	1.078	ns	***	ns
ADF	31.50 ^{BC}	30.53 ^{BC}	37.85 ^{A}	32.02 ^{BC}	34.85 ^{AB}	34.24 ^{AB}	27.99 ^{C}	34.12 ^{AB}	34.91 ^{AB}	0.389	ns	***	*
Ash	13.53 ^{C}	37.85 ^{A}	20.32 ^{AB}	14.18 ^{C}	15.72 ^{BC}	24.73 ^{A}	13.21 ^{C}	19.91 ^{AB}	24.59 ^{A}	0.552	ns	***	ns

$^{A, B, C, D}$Means followed by a different letter within the same row are significantly different: *P < 0.05; **P < 0.01; ***P < 0.001, ns: not significant difference (P>0.05), SEM: standard error of mean.

Table 3.1.2 Effect of species (S) and cutting date (D, days of regrowth) on DM yield (ton/ha) and chemical composition (% on DM basis) in tropical legumes.

Item	Thapra stylo			Verano stylo			Cavalcade			SEM	S	D	S × D
	30 D	45 D	60 D	30 D	45 D	60 D	30 D	45 D	60 D				
DM yield	2.90^{D}	5.46^{B}	8.07^{A}	4.06^{CD}	4.32^{BC}	7.42^{A}	0.65^{E}	0.94^{E}	-	0.118	***	***	ns
DM	23.14^{B}	24.48^{B}	22.15^{B}	28.60^{AB}	29.42^{AB}	32.56^{A}	28.32^{AB}	34.29^{A}	-	0.594	***	ns	ns
OM	90.48^{A}	88.71^{AB}	82.25^{CD}	87.42^{ABC}	83.66^{BCD}	75.75^{E}	86.08^{ABCD}	80.52^{DE}	-	0.546	*	***	ns
CP	19.69^{A}	17.65^{AB}	14.09^{C}	18.92^{AB}	17.98^{AB}	14.68^{C}	19.25^{AB}	17.46^{B}	-	0.218	ns	***	ns
EE	2.63^{AB}	1.89^{CD}	1.72^{CD}	3.15^{A}	1.87^{CD}	1.74^{CD}	2.32^{BC}	1.52^{D}	-	0.071	ns	***	ns
NDF	46.86^{B}	54.95^{AB}	59.57^{A}	44.60^{B}	52.00^{AB}	54.38^{AB}	53.05^{AB}	53.84^{AB}	-	0.835	ns	**	ns
ADF	27.62^{C}	33.77^{AB}	36.16^{A}	30.59^{BC}	37.30^{A}	36.42^{A}	35.88^{AB}	33.29^{AB}	-	0.471	ns	**	*
Ash	9.68^{E}	11.29^{DE}	17.75^{BC}	12.58^{CDE}	16.34^{BCD}	24.25^{A}	15.54^{BCD}	19.48^{AB}	-	0.537	***	***	ns

$^{A, B, C, D, E}$ Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001; ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested.

Table 3.1.3 Effect of species (S) and cutting date (D, days of regrowth) on fatty acid composition (g/100 g total fat) in the tropical grasses.

Item	Purple guineagrass			Mulato II grass			Napiergrass			SEM	S	D	S × D
	30 D	45 D	60 D	30 D	45 D	60 D	30 D	45 D	60 D				
C12:0	0.79 ^{AB}	0.91 ^{A}	0.77 ^{AB}	0.44 ^{BC}	0.71 ^{AB}	0.08 ^{D}	0.27 ^{CD}	0.45 ^{BC}	0.87 ^{A}	0.026	**	ns	**
C14:0	1.07 ^{C}	1.11 ^{C}	1.63 ^{AB}	1.53 ^{ABC}	1.32 ^{BC}	1.31 ^{BC}	0.34 ^{D}	1.86 ^{A}	1.27 ^{BC}	0.035	ns	**	***
C15:0	0.89 ^{B}	0.62 ^{B}	0.71 ^{B}	0.87 ^{B}	0.63 ^{B}	0.02 ^{C}	0.20 ^{C}	1.68 ^{A}	0.02 ^{C}	0.021	*	***	***
C16:0	19.94 ^{A}	16.73 ^{C}	17.03 ^{C}	20.28 ^{A}	17.71 ^{BC}	17.26 ^{C}	20.84 ^{A}	19.83 ^{A}	19.40 ^{AB}	0.138	***	***	ns
C16:1	2.14 ^{A}	2.14 ^{A}	1.48 ^{B}	1.43 ^{B}	0.97 ^{BC}	0.34 ^{DE}	0.17 ^{E}	0.83 ^{CD}	1.48 ^{B}	0.045	***	ns	***
C17:0	0.84 ^{ABC}	0.93 ^{AB}	1.02 ^{A}	0.70 ^{BCD}	0.48 ^{D}	0.03 ^{E}	0.19 ^{E}	0.65 ^{CD}	0.05 ^{E}	0.017	***	**	***
C18:0	4.92 ^{A}	5.63 ^{A}	5.84 ^{A}	2.40 ^{BC}	3.38 ^{B}	3.20 ^{B}	1.74 ^{C}	2.55 ^{BC}	2.58 ^{BC}	0.089	***	*	ns
C18:1n9	14.74 ^{BC}	14.71 ^{BC}	15.33 ^{B}	12.16 ^{D}	18.32 ^{A}	18.99 ^{A}	12.42 ^{D}	12.85 ^{CD}	13.23 ^{CD}	0.148	***	***	***
C18:2n6	12.08 ^{E}	22.14 ^{BCD}	22.43 ^{BC}	28.40 ^{A}	27.21 ^{AB}	30.48 ^{A}	17.16 ^{D}	19.39 ^{BC}	22.81 ^{BC}	0.511	***	**	*
C18:3n3	35.66 ^{B}	28.23 ^{D}	28.18 ^{D}	28.25 ^{D}	21.49 ^{E}	15.59 ^{F}	40.31 ^{A}	33.83 ^{BC}	30.58 ^{CD}	0.469	***	***	ns

$^{A-F}$Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

Table 3.1.3 (Cont.) Effect of species (S) and cutting date (D, days of regrowth) on fatty acid composition (g/100 g total fat) in the tropical grasses.

Item	Purple guineagrass			Mulato II grass			Napiergrass			SEM	S	D	S × D
	30 D	45 D	60 D	30 D	45 D	60 D	30 D	45 D	60 D				
SFA	28.45 ^{A}	25.93 ^{AB}	27.68 ^{A}	26.22 ^{AB}	24.24 ^{BC}	21.92 ^{C}	23.59 ^{BC}	27.03 ^{A}	24.06 ^{BC}	0.190	***	ns	**
MUFA	16.87 ^{B}	16.86 ^{B}	16.51 ^{B}	13.59 ^{CD}	19.29 ^{A}	19.32 ^{A}	12.59 ^{D}	13.68 ^{CD}	14.71 ^{C}	0.138	***	***	***
PUFA	47.74 ^{BC}	50.37 ^{ABC}	50.61 ^{ABC}	56.65 ^{AB}	48.70 ^{BC}	46.06 ^{C}	59.30 ^{A}	53.22 ^{ABC}	51.74 ^{ABC}	0.889	*	ns	*
PUFA/SFA	1.69 ^{C}	1.94 ^{BC}	1.91 ^{BC}	2.24 ^{AB}	2.05 ^{BC}	2.01	2.51 ^{A}	1.97 ^{BC}	2.09 ^{AB}	0.041	**		ns

$^{A-F}$Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

For legume forage, the proportion of C16:0 and C18:1n9 had no influence of legume species, whilst there were significant difference ($P<0.001$) for the proportion of C18:0, C18:2n6 and C18:3n3. The C16:0 content decreased with higher cutting date for the Verano stylo, but only decreased values of the C16:0 contents in the Thapra stylo and the Calvalcade were found small decrease with higher cutting date. There was no systematic pattern for the C18:0, C18:1n9, C18:2n6 and C18:3n3 contents for the studied legumes. The factor of cutting date was found highly significant difference ($P<0.01$) for the proportion of C16:0 and C18:0 and very highly significant difference ($P<0.001$) for the contents of C18:1n9 and C18:2n6, while no effect of days of regrowth and interaction for C18:3n3. The content of C16:0 decreased with higher cutting date. The proportion of C18:0 was no systemic pattern with cutting date increased. The C18:1n9 proportion decreased with higher cutting date whiles the C18:2n6 content increased with more cutting date, except for Cavalcade. The content of C18:3n3 was no systemic pattern with cutting date increased. For grouped FAs, the SFA content decreased with higher cutting date, whereas the proportion of PUFA increased with the cutting date. The ratio of PUFA/SFA for the Mulato II increased and highest at 60 days after regrowth. The ratios of PUFA/SFA were more than 1 (1.15 - 2.54).

Table 3.1.4 Effect of species (S) and cutting date (D, days of regrowth) on fatty acid composition (g/100 g total fat) in the tropical legume.

Item	Thapra stylo			Verano stylo			Cavalcade			SEM	S	D	S × D
	30 D	45 D	60 D	30 D	45 D	60 D	30 D	45 D	60 D				
C12:0	0.25 ^{B}	0.35 ^{B}	0.92 ^{A}	0.26 ^{B}	0.39 ^{B}	0.97 ^{A}	0.16 ^{B}	0.34 ^{B}	-	0.027	**	***	ns
C14:0	0.98 ^{A}	1.05 ^{A}	0.84 ^{A}	1.01 ^{A}	1.35 ^{A}	0.90 ^{A}	1.05 ^{A}	1.10 ^{A}	-	0.043	ns	ns	ns
C15:0	0.67 ^{AB}	0.50 ^{B}	0.03 ^{C}	0.49 ^{B}	0.74 ^{AB}	0.07 ^{C}	0.17 ^{C}	0.80 ^{A}	-	0.019	ns	***	**
C16:0	26.24 ^{A}	26.38 ^{A}	24.15 ^{AB}	26.99 ^{A}	26.67 ^{A}	20.34 ^{B}	25.51 ^{A}	24.50 ^{AB}	-	0.360	ns	**	ns
C16:1	1.54 ^{BC}	2.83 ^{A}	1.19 ^{C}	0.51 ^{D}	1.49 ^{BC}	1.62 ^{BC}	1.41 ^{BC}	1.91 ^{B}	-	0.044	**	***	***
C17:0	0.64 ^{A}	0.50 ^{A}	0.39 ^{A}	0.40 ^{A}	0.43 ^{A}	0.49 ^{A}	0.58 ^{A}	0.59 ^{A}	-	0.043	ns	ns	ns
C18:0	6.01 ^{BC}	4.12 ^{F}	5.38 ^{CDE}	4.79 ^{DEF}	4.49 ^{EF}	5.53 ^{BCD}	7.12 ^{A}	6.44 ^{AB}	-	0.074	***	**	ns
C18:1n9	7.63 ^{A}	6.38 ^{AB}	4.75 ^{D}	5.95 ^{B}	5.82 ^{BC}	4.85 ^{C}	6.54 ^{AB}	7.21 ^{A}	-	0.085	ns	***	***
C18:2n6	15.69 ^{B}	18.07 ^{B}	26.89 ^{A}	15.58 ^{B}	14.45 ^{B}	18.32 ^{B}	23.18 ^{A}	23.78 ^{A}	-	0.499	***	***	*
C18:3n3	26.46 ^{C}	28.76 ^{C}	29.94 ^{C}	37.77 ^{AB}	39.09 ^{A}	44.65 ^{A}	30.80 ^{BC}	31.20 ^{BC}	-	0.744	***	ns	ns

$^{A-F}$Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

Table 3.1.4 (Cont.) Effect of species (S) and cutting date (D, days of regrowth) on fatty acid composition (g/100 g total fat) in the tropical legume.

Item	Thapra stylo			Verano stylo			Cavalcade			SEM	S	D	S × D
	30 D	45 D	60 D	30 D	45 D	60 D	30 D	45 D	60 D				
SFA	35.48 ^{A}	32.90 ^{AB}	30.45 ^{BC}	34.00 ^{AB}	34.08 ^{AB}	27.60 ^{C}	34.60 ^{AB}	34.00 ^{AB}	-	0.330	ns	**	*
MUFA	9.32 ^{A}	9.21 ^{A}	4.18 ^{D}	6.45 ^{C}	7.31 ^{B}	6.47 ^{C}	7.95 ^{B}	9.12 ^{A}	-	0.064	**	***	***
PUFA	40.82 ^{D}	46.82 ^{CD}	56.83 ^{B}	53.35 ^{B}	53.53 ^{BC}	66.57 ^{A}	53.98 ^{BC}	54.98 ^{B}	-	0.744	**	***	ns
PUFA/SFA	1.15 ^{D}	1.59 ^{CD}	1.97 ^{B}	1.62 ^{BC}	1.57 ^{BC}	2.54 ^{A}	1.61 ^{BC}	1.71 ^{BC}	-	0.048	*	***	ns

$^{A-F}$Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

3.1.6 Discussion

3.1.6.1 DM yield and chemical compositions

The current study results had clearly shown that cutting date had influence on the DM yield of the forage. The increase in DM yield of all studied forage accompanied by the cutting date increased was consistent with Elgersma et al. (2003); Vanhato et al. (2007); Grabber (2009) and Abbiasi et al. (2012). Possible demonstrations for the observed responses to the cutting date for the more mature forage would be the increases of leaf and photosynthesis, thus resulting in higher DM yield production with more contents of NDF and ADF in forage. Man and Wiktorsson (2003) studied in Elephantgrass and Guineagrass recorded a similar effect which DM yield in the grasses increased as cutting dates increased (4, 6, 8 and 10 week). Similar results were obtained comparing the 3, 4, 5, 6, 8 and 10 weeks cutting date, DM yield obtained both Guineagrass and Elephantgrass at 10 weeks was higher than that obtained under any of the other intervals (Omaliko, 1980). For Purple guineagrass in the present study, the DM yield was closed to the report earlier (Hare et al., 2009). In addition, the DM yields in the grass deem to be higher than that of legume in accordance with Sengul (2003); Lithourgidis et al. (2006) and Marley et al. (2005). Barnes and Addo-Kwafo (1996) also found that the DM yields in legume forage (*Centrosema pascuorum, Stylosanthes guianensis, Stylosanthes hamata*) and grass forage *(Panicum maximum* T58 and *Brachiaria brizantha)* in almost all treatments increased as regrowth period lengthened (3 to 6 weeks). They also found that grass forage had higher DM yield than legume forage, moreover, DM yields in dry season were higher than wet season, except for Cavalcade (*Centrosema pascuorum)* had DM yield in wet season higher than dry season. In the current study, the Cavalcade may

have similar responses to the effects of dry season, resulting in not enough Cavalcade to be harvested.

The increase of DM yield with cutting date was similar to the higher content of DM, fiber compounds (ADF and NDF) and ash, while the content of CP and EE were reduced. These results would indicate the decrease of forage quality with age as previously reported by Chaves et al. (2006); Fulkerson et al. (2007) and Sengul (2003). In addition, Deinum and Dirven (1972) reported a longer growing period (2, 3, 4 and 5 weeks) in Congograss (*Brachiaria ruziziensis*) showed a decrease in percentage of organic nitrogen and ash, and an increase in percentage of crude fiber. Man and Wiktorsson (2003) reported that the decline in CP concentration in older forage and digestibility of DM and CP decreased as cutting interval increased, indicating lower nutrient availability. Adjei and Fianu (1985) found that the forage legumes decrease in average contents of CP from 22.5 to 17.5% for leaves and 11.9 to 9.4% for stem with longer cutting interval (60, 90 and 120 days after regrowth) and the accompanying increase in average content of CF from 20.0 to 26.8% for leaves and 27.1 to 31.9% for stem. This would imply that the changes of nutrient contents of both legumes and grasses would be the results of the decline or the increase of nutrients in both leaves and stem. Herrero et al. (2001) reported that structural constituents (such as NDF, ADF, cellulose and $KMnO_4$ lignin) had strongest correlations with shear strength in four species of *Brachia*. Therefore, this implies that an increase of structural constituents in forage would partly affect to physical processes (mastication, rumination, microbial degradation and detritions) for particle-size reduction in the ruminants. These results indicated that optimal DM yield with low structural constituents would be considered for choosing forage for the ruminants.

Among three species of the studied grasses, the Multo II grass would be suitable grass forage for the ruminants as giving high DM yield, high CP and low NDF, especially the Mulato II grass harvested at the 45 days after regrowth. For the experimental legume species, the DM yield of the Thapra stylo and the Verano stylo was predominantly higher than that of the Cavalcade, while the chemical composition of the legumes are closed to each other for all three legumes. From the study of Winter et al. (1989), the *Stylosanthes hamata* could be grew reasonably well in soils of low fertility. In addition, the study of Cesar et al. (1999) reported that pasture management by mixed *Panicum maximum* plus *Stylosanthes hamata* was rather was superior to natural grasslands as the results of more resistant to close grazing for sward and providing better nutritive value. Thus, the well grew in low fertility soils and resistance to grazing for the Verano stylo would be the reasons for choosing it as legume forage for the ruminants when compared with the Thapra stylo and Cavalcade.

The large differences of nutrient contents between the forage grasses and legumes of the present study was that the grass contained low CP and high NDF whereas the legume had high CP and low NDF in accordance to the reports of Vanhatalo et al., 2007 and Grabber, 2009. This pattern of differences is also similar to the results reported by Bamikole et al. (2001) who established that Verano Stylo had a higher CP concentration than Guineagrass, and lower concentration of NDF than Guineagrass at 6 weeks harvested. This assertion was supported by the finding of Bamikole et al. (2004) who found that the Verano Stylo had a higher concentration of OM and CP, and lower concentration of NDF than Guineagrass at every 45 day harvested. Furthermore, Kanani et al. (2006) reported that the forage legumes had over 20% CP, while Sudangrass contained only 7.8% CP, which had high fiber content.

These would make the conclusion for the current study that the studied grasses contain high NDF and low CP while the experimental legumes contain low NDF and high CP. As mentioned earlier, grass was better than legume for giving higher DM yield. Hence, both types of forage would be selected for the ruminants, but the balance between DM yield and chemical composition would be considered for optimal forage properties for the ruminants under conditions of the current study.

3.1.6.2 Fatty acid compositions

Data from the current study have demonstrated that the grasses and the legumes vary in their FA profiles. Both types of forage contained a large proportion of C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3, when compared with the other FAs. These results were in consistent with those reported by Dewhurst et al. (2001); Clapham et al. (2005), Vanhatalo et al. (2007) and Mel'uchová et al. (2008) who found predominant proportion of C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3 in grasses and legumes.

In the current study, the forages with advances in the maturity were associated with fluctuation of proportion of FAs. The concentration of C16:0 for both experimental grasses and legumes were the most abundant proportion in SFA and were decreased with higher cutting date after regrowth. The studied grasses had lower proportion of C16:0 (16.7-20.8%) than the legumes (20.3-27.0%). The current study found the proportions of C18:0 increased for the grasses, but tended to decrease for the legumes, when the cutting date was higher. Dewhurst et al. (2001) and Elgersma et al. (2003) also reported the ryegrass reduced the content of C18:0 with increased cutting date. However, Clapham et al. (2005) observed decreased in the concentration of C18:0 for both forages (grass and legume) which was in agreement with the report

of Vanhatalo et al. (2007). The concentrations of C18:1n9 from the grasses tended to elevate with increasing maturity whereas the legumes had a trend of lower concentration of C18:1n9 with advancing maturity, except for the studied Calvacade. This was in accordance with the report of Vanhatalo et al. (2007) who found that timothygrass (*Phleum pratense*) had increased C18:1n9 at the late growth of cutting, while red clover (*Trifolium protense*) contained lower C18:1n9 content with advancing maturity. Similarly, Clapham et al. (2005) reported that the concentration of C18:1n9 decreased in orchardgrass and perenial ryegrass with 3-week harvested interval. They also found that the white clover, as plant grew (3-week interval of regrowth) had lower proportion of C18:1n9. From the study of Jaturasitha et al. (2009), the Purple guineagrass and the Thapra stylo legume had closed concentration of C16:0 each other (21.02 and 22.23% of total analyzed FA, respectively). In the meantime, Dewhurst et al. (2001) reported that the content of C16:0 and C18:1n9 in ryegrasses (*Lolium perenne* and *L. multiflorum*) harvested at 20 days after regrowth (younger) were higher than those harvested at 38 days after regrowth (older). The experimental grasses had a trend of lower C18:3n3 proportion associated with advancing maturity, while the proportions of C18:2n6 were tended to increase with the higher cutting date. A positive relationship was found between cutting date and C18:2n6 in the legumes and no effect on C18:3n3 in the all studied legumes. In the study of Clapham et al. (2005), it was found that the white clover, orchardgrass and perennial ryegrass have a higher proportion of C18:3n3 and a lower proportion of C18:2n6, and concentrations declined in all plant materials as plant grew. Elgersma et al. (2003) also noted that a loss of C18:3n3 in fresh grass (perennial ryegrass) between 23 and 33 days after regrowth, in agreement with the results of Dewhurst et al. (2002). In the study of

Jaturasitha et al. (2009), Guineagrass had lower C18:2n6 content than Thapra stylo (16.73 and 19.50% of total analyzed FA, respectively), whereas Guineagrass had higher C18:3n3 content than Thapra stylo (48.94 and 44.68% of total analyzed FA, respectively). Indeed, C18:3n3 concentration reaching much higher than our experiment, suggesting that the variation in the α-linolenic acid concentration in forage appeared to be associated with environment such as solar radiation and temperature (Witkowska et al., 2008) in accordance with the report of Hawke (1973) who reported the concentration of C18:3n3 and TFA were higher at lower temperature. Moreover, cutting and drying of forages may cause significant reductions in FA content and percentage of USFAs (Barnes et al., 2007). In the reports mentioned earlier, there were different directions of FA contents in both forage and legume forages. From the results of the current study, it has been shown effect of species of grasses and legumes, cutting date and their interaction on FA profile. This would explain for variation of change of FA concentration in the forage.

Additionally, this study revealed that comparison of the FA composition of forage (grass and legume) indicated both advances in the stage of maturity, there were variation of SFA, MUFA and PUFA. The study of Vanhatalo et al. (2007) reported lower SFA, but increased MUFA and PUFA with forage maturity. In the mean time, Clapham et al. (2005) and Boufaïed et al. (2003) found that in grasses (orchardgrass and perennial ryegrass) and legume (white clover) decreased the concentration of SFA, MUFA and PUFA with plant maturity. From the reports mentioned earlier, the variations in FA profile coincided with differences in forage development stage (Witkowska et al., 2008). The total of main FAs in each cutting date of both grasses and legumes was a little different, indicating an increase of some FAs at the expense

of the other FAs.

In the first part of the present study, there was an increase of structural constituents; NDF, ADF and ash, whilst the OM, CP and EE, decreased for both grasses and legumes. This would be attributed to dilution effects of growth and increased concentrations of other metabolites, such as cellulose, hemicellulose and lignin (Clapham et al., 2005). The current study found the obviously decrease of the EE content in both grasses and legumes, leading to change in proportion of FAs the forage resulted in varied change pattern of FA profile in the forage. This also was in agreement with the study of Khan et al. (2012) who found the positive relationship between the total fat contents and key FA concentrations (C16:0, C18:1, C18:2 and C18:3) in silages. For choosing a suitable grass and legume for the ruminants by considering FA profile, there was quite difficult for the current study as dynamic change of FA profile. However, the key point for all studied forage species was the results that almost FAs of the forage at 45 days after regrowth was in between that of the forage at 30 and 60 days after regrowth. This implied the possibility to choose cutting date at 45 days after regrowth of all studied forage for the ruminants. When considered chemical composition and properties of forage together with FA profile, the suitable forage species and cutting date of the experimental forage for the ruminants were Mulato II grass and Verano stylo with the harvesting at 45 days after regrowth.

3.1.7 Conclusions

The experimental grasses (Purple guineagrass, Mulato II grass and Napier grass) and legumes (Thapra stylo, Verano stylo and Cavalcade) would increase DM yield and content of DM, NDF, ADF and ash, but decrease contents of OM, CP and

EE with advancing maturity of the forage. However, there were inconsistent pattern of FA profiles in forages, which might be attributed by dilution effects of forage growth and structural constituents. The Mulato II grass and Verano stylo harvested at 45 days after regrowth would contain proper chemical composition and properties including proper FA profiles for ruminants.

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3.2 Experiment I-II : Effect of forage species at different regrowth age and additive on chemical composition, fermentation quality and fatty acid composition of tropical forage silage

3.2.1 Abstract

The current experiments was carried out to investigate the chemical composition and fermentation quality of silage for 6 species of tropical forage at 3 different cutting date (30, 45 and 60 days after regrowth) and also influences of 4 additive treatments on chemical composition, fermentation quality and fatty acid profile. The studied 3 grasses were Purple guineagrass (*Panicum maximum* TD. 58), Mulato II grass (*Brachiara ruziziensis* × *B. brizantha* × *B. decumbens*) and Napiergrass (*Pennisetum purpureum* Schumacher) and the experimental 3 legumes were Verano Stylo (*Stylosanthes hamata*), Thapra Stylo (*Stylosanthes guianensis* CIAT 184) and Cavalcade (*Centrosema pascuorum*). Individual forage species and cutting date were combined with 4 additive treatments (no additive, cassava meal 5%, molasses 2% and the fermented juice of epiphytic lactic acid bacteria (FJLB) 1%). The study design was 3 × 3 × 4 factorial arrangements in CRD for grass and legume, respectively. The samples of the silages were sampled and then prepared for FA analysis by gas chromatography. The important results of the current study have shown that species of grass in the current study had influences on all chemical composition, except for the ADF. There was higher pH, NH_3-N, lactic acid (LA), acetic acid (AA), propionic acid (PA) and total volatile fatty acids (VFAs) but lower butyric acid (BA) for the Purple Guineagrass silage and the Mulato II grass silage, when compared with those of the Napiergrass silage. The chemical composition of

the grass silages with advanced cutting date seemed to be related to the chemistry contents in fresh grasses, but there were inconsistent with the cutting date. There were low pH, NH_3-N and BA, but high in LA, AA, PA and total VFAs for the grass harvested at 45 days after regrowth. The molasses adding for ensiling had clearly lowering NDF content of grass silage while there were inconsistent effects on chemical composition for the other additives. For legume silage, the legume species had influence on all analysis chemical compositions. There was clearly effect of legume species on formation of VFAs. The chemical composition of the legume silages with advanced cutting date was seemed to be related to the chemical profiles for NDF and ADF in fresh legumes, while the final legume silages with advancing cutting date had a trend of higher CP and EE; especially the CP contents at 45 days after regrowth and EE contents at 45 and 60 days after regrowth. There was rather evident that the legumes supplemented with each additive improved fermentation quality compared with the legumes without additive supplementation as depressing pH value, NH_3-N and BA whereas increasing LA and AA. The FA profile of the experimental grass silages composed main FAs; C16:0 (15.62-22.86%), C18:0 (0.54-2.24%), C18:1n9 (1.67-3.97%), C18:2n6 (13.12-17.23%) and C18:3n3 (41.57-51.42%), while the rest of the analyzed FAs were lower than 2% of total fat for all treatments. The main FAs of the legume silages were C16:0 (14.79-22.77%), C18:0 (1.50-5.42%), C18:1n9 (1.27-4.66%), C18:2n6 (16.38-25.98%) and C18:3n3 (35.00-45.02%), while the rest of the analyzed FAs were lower than 2% of total fat for all treatments of the legume silages. There were small numerical changes of individual FA contents in all experiment treatments. In conclusion, the studied additives (cassava meal, molasses and FJLB) would preserve FA contents of grass and legume silages.

In conclusion, the Mulato II grass and the Verano stylo, harvested at 45 days after regrowth were suitable forage for making silage for goats as their chemical composition and fermentation quality of the final silages. Adding FJLB into Mulato II grass and Verano stylo would beneficial for farmers as an alternative additive of low cost and easy preparing.

Key Words: Cutting date, Additive, Chemical compositions, Fermentation quality, Fatty acid composition, Tropical forage silage

3.2.2 Introduction

Forages, such as grass and legume, are preserved as silage, especially during dry season. The silage can be provided as feed for ruminant production in dry season by supplementing the diet with a valuable source of energy and protein (Heinritz et al., 2012). In principle, forage silage was made by controlled anaerobic fermentation. An important technique to make good silage is using the external weight to squeeze out all of the air from contained bag and arresting the natural process of oxidation and decay of harvested forages. Silage is produced successfully when bacteria producing LA dominate fermentation and restrict the activity of clostridia (Bureenok et al., 2006). There are quite difficult to make silage and to control fermentation quality for tropical forage silage, because tropical forage are low in WSC, high buffering capacity and low LAB (Catchpoole and Henzell, 1971, and Niimi and Kawamura, 1998). Most of grasses have high moisture content and low soluble carbohydrate levels (Nussio, 2005) whereas most of legumes have low sugar content and high buffering capacity (McDonald, 1991). Many researchers have attempted to devise for improving

the quality of tropical forage silage. Supplementation of additive for ensiling is usually way for the improvement. Increasing supply of WSC for ensiling resulted in producing sufficient LA for rapid pH reduction and improving the fermentative quality of silage made from tropical forage as the reports earlier (Bureenok et al., 2005a; Yahaya et al., 2004; Tamada et al., 1999; Sibanda et al., 1997). The additives commonly used for tropical forage ensiling are cassava meal, molasses and LAB.

It is a well-known fact that LAB plays a crucial role in silage fermentation (Bureenok et al., 2005b). Lactic acid bacteria are generally added into forage to enhance the nutritional value of silage and prevent the growth of fungi or yeast that could cause aerobic spoilage (Amado et al., 2012; Flythe and Russell, 2004; Woolford, 1990). In studies earlier, inoculation of LAB at silage has been completed by many groups with inconsistent results. Sometime it was effective (Kumai et al., 1990; Tengerdy et al. 1991; Masuko et al., 1992; Rooke and Kafilzadeh, 1994) whilst sometime it was not (Lindgren et al., 1983). An important factor affecting the success of ensiling is a number of species and strains of LAB applied, which is related to adaptation to the specific environment and enhancing the LA production (Ohshima et al. 1997).

The natural microorganisms presented in forage crops are responsible for fermentation of silage and influence quality of silage. In addition, the proportional population of LAB is usually low and variable with standing crops (Muck 1990, and Lin et al., 1992). There are many reports showing that fermented juice of epiphytic lactic acid bacteria (FJLB), a culture solution produced by LAB, have been used successfully to improve the nutritive value of various silage preparations (Ohshima et al., 1997; Masuko et al., 2002; Bureenok et al., 2005a, Takahashi et al., 2005;

Horiguchi and Takahashi, 2007), for alfalfa, Timothy and Orchardgrass, Guineagrass, rice, green soybean stover. However, there is no information available for ensiling of Purple guineagrass, Mulato II grass and Napiergrass and a little information for Thapra stylo, Verano stylo and cavalcade silages. All six forages mentioned earlier are tropical forage found in Thailand and are used for ruminants. Ensiling for both forage species would be an alternative preservation during dry season of shortage of forage.

The aim of the present study was to determine the effect of FJLB, cassava meal and molasses as the additive treatment in the tropical forage harvested at 30, 45 and 60 days after regrowth on the nutritive value, silage quality and FA compositions.

3.2.3 Materials and methods

3.2.3.1 Plant materials

Forages used in the study were three species of grasses: Purple guineagrass (*Panicum maximum* TD. 58), Mulato II grass (*Brachiara ruziziensis* × *B. brizantha* × *B. decumbens*) and Napiergrass (*Pennisetum purpureum* Schumacher) and three legumes: Verano stylo (*Stylosanthes hamata*), Thapra stylo (*Stylosanthes guianensis* CIAT 184) and Cavalcade (*Centrosema pascuorum*). A series of 30 plots (each 3 m × 3 m) was sown without fertilizer on February 2008 at Faculty of Natural Resources, Rajamangala University of Technology-Isan, Sakon Nakhon Campus (located in North-Eastern of Thailand). Forage samples were taken in May, July and September 2008 at 30, 45 and 60 days after regrowth.

3.2.3.2 Fermented juice of epiphytic lactic acid bacteria (FJLB) preparation

The FJLB was prepared from Purple guineagrass, Mulato II grass,

Napiergrass, Thapra stylo, Verano stylo and Cavalcade before harvesting; 200 g of fresh grass was macerated with 600 ml of distilled water in a blender. The macerate was filtered and 50 ml of the filtrate was put into a flask. These filtrates in the flask was treated with glucose at the rate of 2% of volume and incubated at 30 °C for 2 days.

3.2.3.3 Silage making

After forage harvesting, the experimental forage were immediately chopped into 1-2 cm-length pieces. Then, cassava meal, molasses, and FJLB were added at 2, 5 and 10% of fresh matter as a silage additive, respectively while no additive was added for the control grass and legume silages. Five replicated plastic bags per each treatment were prepared and allowed to ferment for 80 days at room temperatures.

3.2.3.4 Chemical analysis

Dry matter content of the fresh materials and silages were determined by drying in a hot-air oven at 60 °C for 72 hours, then ground to pass through a 1 mm mesh screen and subsequently analyzed for chemical composition. Total N was determined using the Kjeldahl method and crude protein (CP) was calculated by multiplying the N content by 6.25. Ether extract (EE) and ash contents were quantified by AOAC (1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) determined by the methods described by Goering and van Soest (1970).

3.2.3.5 Fermentation quality evaluation

After each bag of Purple guineagrass, Mulato II grass, Napiergrass, Thapra stylo, Verano stylo and Cavalcade silages was opened, the silage content was mixed thoroughly. Then, 20 g of the content was sampled from each bag and followed by adding about 70 g of distilled water and then macerating at 4 °C for 12 h and

subsequently all samples were measured pH values by using a glass electrode pH meter. These sample solutions were filtered through two layers of cheesecloth and a filter paper no. 1, and then the filtrate was stored at -20 °C prior to chemical analysis. The filtrate was used for determining ammonia nitrogen (NH_3-N) (by the Kjeldahl method), lactic acid (LA), acetic acid (AA), propionic acid (PA) and butyric acid (BA). The LA, AA, PA, BA and total volatile fatty acids (VFAs) were determined by high performance liquid chromatography (HPLC, Shim-pack SCR-102H, 300 mm × 8.0 mm i.d.; column temperature, 40 °C; flow rate, 0.8 ml/min, Shimadzu Co. Ltd., Kyoto, Japan).

3.2.3.6 Fatty acid methyl ester of oil samples

After each bag of Purple guineagrass, Mulato II grass, Napiergrass, Thapra stylo, Verano stylo and Cavalcade silages were opened, the silage content was mixed thoroughly. Then, 20 g of the content was sampled from each bag. Each sample was followed by adding about 70 g of distilled water and then macerating at 4 °C for 24 h and subsequently these sample solutions were filtered through two layers of cheesecloth and a filter paper (no. 1). The filtrate was stored at -20 °C and subsequently freeze-dried and prepared for FA analysis by gas chromatography (GC) of fatty acid methyl ester (FAME). The lipid was extracted from the forage using the chloroform/methanol (2/1) method procedure of Folch et al. (1957) and methylation of sample by the procedure described by Metcalfe (1966) was used. Fatty acid composition was measured after methylation of samples. Fatty acid methyl esters were analyzed on a Perkin Elmer Auto system gas chromatograph equipped with a flame-ionization detector (FID) using a capillary column (SPTM-2560, 100 m × 0.25 mm ID, 0.20 μm film). This analyzed adopted a split injection (split ratio 100:1).

The GC analysis was temperature programmed, at 140 °C held at 5 min, and raise from 140 °C to 240 °C at a rate of 4 °C/min and then held 240 °C for 40 min. The injection port and detector temperatures were set at 260 °C. Helium was used as the carrier a gas at a rate of 20 cm/sec. Identification of the FA was based upon retention times using standards of methyl esters. A mixture of the standards of the individual FAME was used to determine response factors. The areas of the peaks in the chromatogram were calculated and normalized using response factors. Proportions of FA were calculated.

3.2.4 Statistical analysis

3.2.4.1 Chemical compositions and fermentation quality

Data were statistically analyzed according to 3 × 3 × 4 factorial arrangements in CRD using the PROC GLM procedure (SAS, 1990). Significant differences ($P<0.05$) among treatments were determined using Duncan's News Multiple Range Test according to Steel and Torrie (1980).

3.2.4.2 Fatty acid compositions

Data were statistically analyzed according to 3 × 4 factorial arrangements in CRD using the PROC GLM procedure (SAS, 1990). Significant differences ($P<0.05$) among treatments were determined using Duncan's News Multiple Range Test according to Steel and Torrie (1980).

3.2.5 Results

3.2.5.1 Chemical composition

Chemical composition of grass silage has shown in Table 3.2.1. The DM content of grass silage had significant influences of species; S ($P<0.001$), cutting date;

D ($P<0.05$) and additive; A ($P<0.01$) with no presentation of all interactions. The OM concentration of the grass silage had significant influences of species ($P<0.001$), day after regrowth ($P<0.001$) and interaction between species (S) and cutting date (D) ($P<0.01$) with no effect of additive and no presentation of the interactions of S × A, D × A and S × D × A. The CP proportion in the grass silage had significant influences of species ($P<0.001$), day after regrowth ($P<0.001$) and additive ($P<0.01$) with presentation of S × A ($P<0.05$), D × A ($P<0.001$) and S × D × A ($P<0.001$), but no interaction of S × D. The grass silage had effect of all factors (S, D and A) ($P<0.001$) and all interadions ($P<0.001$) on the EE content. For the NDF in grass silage had significant influences of species ($P<0.01$), cutting date ($P<0.01$) and additive ($P<0.05$) with significant interaction of D × A ($P<0.01$), but no interaction of S × D, S × A and S × D × A. The factors of cutting date and interaction of D × A influenced ($P<0.001$) on the ADF content of grass silage, but no effect of species, additive and interaction of S × D, S × A and S × D × A. For ash content in grass silage, there were presented influence of species ($P<0.001$), cutting date ($P<0.05$) and interactions of S × D ($P<0.001$) and S × A ($P<0.05$), without effect of additive and interaction of D × A and S × D × A.

For legume silage, chemical composition has shown in Table 3.2.2. The DM content of legume silage had significant influence on species ($P<0.001$) and cutting date ($P<0.01$) with no effect of additive and no presentation of all interactions. There was only effect of species ($P<0.01$) on the OM concentration in the legume silage, the rest of test factors and all interactions had no influence. The CP proportion in the legume silage had significant influence on species ($P<0.001$), cutting date ($P<0.001$) and interaction of S × D ($P<0.001$), S × A ($P<0.001$), and S × D × A

(P<0.01), with no effect of additive and interaction of D × A. The EE proportion in the legume silage had significant influences of species (P<0.001), cutting date (P<0.001) and interaction of S × D (P<0.001), S × A (P<0.001), D × A (P<0.05) and S × D × A (P<0.001), but no effect of additive on the EE content. The NDF content of legume silage had significant influences of species (P<0.05), cutting date (P<0.001) and additive (P<0.001) with no presentation of all interactions. The ADF content of legume silage had significant influence of species (P<0.001), cutting date (P<0.001), additive (P<0.001) and interaction of S × D (P<0.01) with no presentation of the other interactions. There was only effect of species (P<0.05) on the ash content in the legume silage, the rest of test factors and all interactions had no influence.

3.2.5.2 Fermentation quality

Fermentation quality of grass silage is shown in Table 3.2.3. There were effects of grass species, cutting date, additive and interaction of cutting date and additive, while no difference for other interactions was found. The concentration of NH_3-N in the grass silage had significant influence on species (P<0.01), cutting date (P<0.001) and interaction of S × D (P<0.05), D × A (P<0.01) and S × D × A (P<0.01), whereas no influence on additive and interaction of S × A. There were significant effects of contents of LA, AA, PA, BA and total VFAs in grass silage at P<0.001, except for interaction of S × D (P<0.01) for LA and interaction of S × A (P<0.05) for total VFAs.

The legume silage quality of fermentation as showed in Table 3.2.4. The pH of the legume silage was influenced by additive (P<0.001) and interaction of S × D (P<0.05), while there were no influence of legume species, cutting date and the other interactions. For the NH_3-N concentration in the legume silage, there were significant

influence of additive ($P<0.01$) and interaction of D × A ($P<0.05$) and S × D × A ($P<0.05$), whilst no effect of legume species, cutting date and interaction of S × D and S × A. The contents of LA in legume silage were influenced by all tested factors at different significance level ($P<0.001$) for species, cutting date, additive and interaction of D × A ($P<0.01$) for interaction of S × A and S × D × A ($P<0.05$) for interaction of S × D. There were significant effects on contents of LA, AA, PA, BA and total VFAs in legume silage at level of $P<0.001$, except for influence of additive on AA content ($P<0.05$) and no effect of additive on total VFAs.

3.2.5.3 Fatty acid composition of grass silages

In Table 3.2.5, the FA profile of the experimental grass silages in this study has been shown. The main FA were C16:0 (15.62-22.86%), C18:0 (0.54-2.24%), C18:1n9 (1.67-3.97%), C18:2n6 (13.12-17.23%) and C18:3n3 (41.57-51.42%), while the rest of the analyzed FAs were lower than 2% of total fat for all treatments of the grass silages. For the grouped FAs, grass silages mainly contained PUFA (56.06-66.77%) while MUFA and SFA contents in grass silages were ranged from 1.97-5.51% and 19.80-27.12%, respectively. The C16:0 concentration was influenced ($P<0.05$) by grass species. Although there was no systematic pattern of the C16:0 contents, the Purple guineagrass silage without additive had the lowest proportion of C16:0 silage treated with FJLB had highest proportion of C16:0. The content of C18:0 was influenced by the interaction between grass species and additive ($P<0.01$), there was quite low content of C18:0 for the FJLB additive added to the Purple guineagrass silage. The grass species ($P<0.001$) and additive ($P<0.05$) factors affected the concentration of C18:1n9. The content of C18:2n6 and C18:3n3 were influenced ($P<0.05$) by additive. The difference of C18:2n6 content was found between

the cassava meal and FJLB additives treated to the Purple guineagrass and the Napiergrass as higher C18:2n6 content for the grasses treated with the FJLB. For the C18:3n3 concentration, the Mulato II grass silage treated with the FJLB had lower than that treated with the cassava meal. The grass species had influenced on concentration on SFA ($P<0.05$), MUFA ($P<0.01$) and PUFA ($P<0.05$), whereas there were presented effects of the additive ($P<0.01$) and the interaction ($P<0.001$) on proportion of MUFA. The ratios of PUFA/SFA of grass silages were not different among all treatments. Values of all ratios of PUFA/SFA were higher than 2.0 (2.07 - 3.28.).

3.2.5.4 Fatty acid composition of legume silages

From Table 3.2.6, the FA composition of the studied legume silages in the current study has been illustrated. The main FAs were C16:0 (14.79-22.77%), C18:0 (1.50-5.42%), C18:1n9 (1.27-4.66%), C18:2n6 (16.38-25.98%) and C18:3n3 (35.00-45.02%), while the rest of the analyzed FAs were lower than 2% of total fat for all treatments of the legume silages. For the grouped FAs, the grass silages mainly contained PUFA (53.61-67.50%) while MUFA and SFA contents in the grass silages were ranged from 1.75-5.51% and 20.62-26.41%, respectively. The C16:0 concentration was effected ($P<0.01$) by legume species. The Cavalcade silage without additive had highest content of C16:0 but the Verano stylo silage had lowest content of C16:0. However, there was no systematic pattern of the C16:0 contents. The content of C18:0 was influenced by the legume species, the Cavalcade silage treated with all types of additive tended to have higher proportion of C18:0 than the other two legumes. The interaction of the legume species and the additive was presented for the concentration of C18:1n9. The contents of C18:2n6 ($P<0.001$) and C18:3n3 ($P<0.05$)

were influenced by legume species. There was no effects ($P>0.05$) of all tested factors for SFA. The additives had influenced ($P<0.05$) on concentration of MUFA, PUFA and ratio of PUFA/SFA, whereas there were only presented effects of the legume species ($P<0.01$) on content of PUFA. The ratios of PUFA/SFA of legume silages were higher than 2.0 (2.16-3.11).

Table 3.2.1 Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental grasses silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
Purple guineagrass	30	Control	22.70^{ABCDEFGHI}	85.83^{AB}	5.46^{EFGHI}	3.81^{GHIJ}	60.53^{ABCDEF}	36.22^{ABCDE}	14.17^{HI}
		Cassava meal	25.13^{ABCDEFG}	84.77^{AB}	4.40^{I}	3.65^{HIJK}	56.74^{ABCDEFG}	29.91^{ABCDEF}	15.23^{GHI}
		Molasses	21.84^{BCDEFGHI}	86.26^{AB}	4.57^{I}	3.77^{GHIJ}	37.92^{DEFG}	26.94^{CDEF}	13.75^{HI}
		FJLB	22.49^{ABCDEFGHI}	84.34^{AB}	8.33^{B}	3.32^{KLM}	40.48^{CDEFG}	22.05^{DEF}	15.66^{GHI}
	45	Control	26.49^{ABCD}	84.14^{AB}	5.18^{FGHI}	3.99^{FGH}	56.61^{ABCDEFG}	37.50^{ABCDE}	15.86^{GHI}
		Cassava meal	28.64^{A}	85.59^{AB}	5.14^{GHI}	2.86^{NO}	54.25^{ABCDEFG}	36.43^{ABCDE}	14.41^{HI}
		Molasses	26.62^{ABC}	84.12^{AB}	5.31^{EFGHI}	3.79^{GHIJ}	59.16^{ABCDEFG}	37.05^{ABCDE}	15.88^{GHI}
		FJLB	25.94^{ABCDE}	83.46^{AB}	4.91^{HI}	4.35^{DEF}	62.10^{ABCDE}	29.01^{ABCDEF}	16.54^{GHI}
	60	Control	22.13^{BCDEFGHI}	84.43^{AB}	5.23^{FGHI}	3.72^{HIJK}	67.64^{AB}	46.37^{A}	15.57^{GHI}
		Cassava meal	25.46^{ABCDEF}	86.54^{A}	5.49^{EFGHI}	3.77^{GHIJ}	65.48^{ABC}	39.33^{ABCD}	13.46^{I}

$^{A, B, C,\ldots, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.1 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental grasses silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
		Molasses	23.97^{ABCDEFGH}	85.89^{AB}	5.47^{EFGHI}	3.64^{HIJK}	67.25^{AB}	44.86^{AB}	14.03^{HI}
		FJLB	26.72^{AB}	85.33^{AB}	5.11^{GHI}	3.87^{GHI}	74.60^{A}	43.87^{ABC}	14.67 ^{HI}
Mulato II grass	30	Control	19.49^{FGHI}	80.00^{BCDEF}	7.19^{BCDE}	4.94^{B}	62.91^{ABCD}	40.45^{ABC}	20.00^{CDEFGH}
		Cassava meal	23.86^{ABCDEFGH}	85.61^{AB}	6.53^{BCDEFGH}	4.89^{BC}	54.46^{ABCDEFG}	32.14^{ABCDEF}	14.40^{HI}
		Molasses	26.71^{AB}	79.90^{BCDEF}	7.11^{BCDEF}	2.96^{MNO}	34.06^{G}	21.34^{EF}	21.26^{CDEFG}
		FJLB	21.13^{BCDEFGHI}	80.11^{ABCDEF}	7.48^{BCD}	2.94^{MNO}	40.98^{CDEFG}	27.89^{BCDEF}	18.66^{DEFGHI}
	45	Control	21.55^{BCDEFGHI}	74.57^{FG}	5.27^{EFGHI}	5.08^{B}	56.33^{ABCDEFG}	35.76^{ABCDE}	25.43^{BC}
		Cassava meal	25.64^{ABCDEF}	81.25^{ABCDE}	6.62^{BCDEFGH}	3.96^{FGH}	51.24^{ABCDEFG}	32.40^{ABCDEF}	18.75^{DEFGHI}
		Molasses	22.23^{BCDEFGHI}	76.68^{EFG}	6.27^{CDEFGHI}	4.18^{EFG}	48.03^{BCDEFG}	36.28^{ABCDE}	23.32^{BCD}
		FJLB	20.77^{BCDEFGHI}	77.08^{CDEFG}	6.72^{BCDEFGH}	5.48^{A}	53.45^{ABCDEFG}	33.20^{ABCDEF}	23.07^{BCDE}

$^{A, B, C,\ldots, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.1 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental grasses silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
	60	Control	19.22^{FGHI}	83.08^{ABC}	5.31^{EFGHI}	5.67^{A}	59.93^{ABCDEF}	42.65^{ABC}	16.92^{FGHI}
		Cassava meal	$22.50^{ABCDEFGHI}$	84.31^{AB}	5.18^{EFGHI}	4.68^{BCD}	$58.55^{ABCDEFG}$	38.44^{ABCDE}	15.69^{GHI}
		Molasses	$22.47^{ABCDEFGHI}$	83.35^{AB}	$6.20^{CDEFGHI}$	4.51^{CDE}	$58.40^{ABCDEFG}$	39.32^{ABCD}	17.15^{EFGHI}
		FJLB	$20.19^{CDEFGHI}$	83.36^{AB}	5.61^{DEFGHI}	4.84^{BC}	$58.34^{ABCDEFG}$	34.68^{ABCDE}	16.02^{GHI}
Napiergrass	30	Control	16.64^{I}	82.93^{ABCD}	12.79^{A}	3.21^{LMN}	$59.28^{ABCDEFG}$	40.19^{ABC}	17.07^{EFGHI}
		Cassava meal	18.73^{GHI}	82.19^{ABCDE}	5.12^{GHI}	3.45^{IJKL}	$50.27^{ABCDEFG}$	21.54^{EF}	17.82^{DEFGHI}
		Molasses	18.97^{GHI}	80.33^{ABCDEF}	5.38^{EFGHI}	3.71^{HIJK}	40.51^{CDEFG}	16.88^{F}	$19.66^{CDEFGHI}$
		FJLB	16.93^{I}	82.11^{ABCDE}	7.99^{BC}	3.40^{JKL}	37.13^{EFG}	33.50^{ABCDEF}	17.31^{EFGHI}
	45	Control	18.99^{GHI}	71.60^{GH}	$6.58^{BCDEFGH}$	3.71^{HIJK}	36.34^{FG}	40.37^{ABC}	28.40^{AB}
		Cassava meal	$21.64^{BCDEFGHI}$	67.45^{H}	5.68^{DEFGHI}	3.08^{LMNO}	49.01^{BCDEFG}	16.62^{F}	32.56^{A}

$^{A, B, C,\ldots,.P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.1 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental grasses silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
		Molasses	20.12^{DEFGHI}	72.90^{G}	$6.14^{CDEFGHI}$	4.40^{DE}	$59.08^{ABCDEFG}$	29.45^{ABCDEF}	27.10^{B}
		FJLB	$20.36^{BCDEFGHI}$	76.93^{DEFG}	6.88^{BCDEFG}	2.74^{OP}	$55.73^{ABCDEFG}$	36.11^{ABCDE}	22.92^{CDEF}
	60	Control	17.90^{HI}	83.51^{AB}	4.99^{GHI}	3.06^{LMNO}	61.02^{ABCDEF}	45.94^{A}	16.49^{GHI}
		Cassava meal	$21.85^{BCDEFGHI}$	85.28^{AB}	5.18^{FGHI}	2.43^{P}	45.11^{BCDEFG}	35.59^{ABCDE}	14.72^{HI}
		Molasses	19.83^{EFGHI}	85.14^{AB}	$6.29^{CDEFGHI}$	2.92^{MNO}	40.22^{CDEFG}	44.15^{ABC}	14.86^{HI}
		FJLB	18.44^{HI}	85.71^{AB}	5.70^{DEFGHI}	2.79^{NOP}	$58.76^{ABCDEFG}$	42.11^{ABC}	14.29^{HI}
S.E.M			0.287	0.230	0.088	0.029	0.824	0.561	0.229
S			***	***	***	***	**	ns	***
D			*	***	***	***	**	***	*
A			**	ns	**	***	*	ns	ns

$^{A, B, C, \ldots, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.1 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental grasses silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
S × D			ns	***	ns	***	ns	ns	***
S × A			ns	ns	*	***	ns	ns	*
D × A			ns	ns	***	***	**	***	ns
S × D × A			ns	ns	***	***	ns	ns	ns

[A, B, C,…., P]Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.2 Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental legume silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
Thapra Stylo	30	Control	26.26^{CDEF}	87.11^{A}	9.90^{GHIJ}	3.02^{LMN}	41.81^{EFGH}	26.74^{IJK}	12.90^{C}
		Cassava meal	24.19^{DEF}	86.36^{AB}	9.05^{IJ}	3.28^{JKLM}	37.44^{GH}	20.41^{L}	13.64^{BC}
		Molasses	24.09^{DEF}	88.23^{A}	9.08^{IJ}	3.42^{IJKL}	36.76^{H}	23.29^{KL}	11.77^{C}
		FJLB	26.23^{CDEF}	85.44^{AB}	10.52^{EFGHIJ}	3.06^{LMN}	43.37^{DEFGH}	23.52^{KL}	14.56^{BC}
	45	Control	20.77^{F}	78.01^{ABC}	12.95^{DE}	3.79^{EFGHI}	48.58^{ABCDEF}	32.81^{GHI}	21.99^{ABC}
		Cassava meal	23.23^{EF}	85.94^{AB}	14.75^{CD}	3.82^{EFGHI}	42.70^{DEFGH}	27.62^{IJK}	14.07^{BC}
		Molasses	23.23^{EF}	86.11^{AB}	16.38^{C}	3.57^{HIJK}	46.81^{BCDEF}	27.63^{IJK}	13.90^{BC}
		FJLB	21.61^{F}	86.66^{A}	15.62^{C}	3.49^{IJKL}	52.60^{ABC}	29.92^{HIJ}	13.34^{C}
	60	Control	19.74^{F}	86.91^{A}	10.82^{EFGHIJ}	3.80^{EFGHI}	55.35^{A}	50.40^{A}	13.09^{C}
		Cassava meal	23.47^{EF}	87.54^{A}	11.79^{EFGHI}	4.57^{ABC}	47.56^{ABCDEF}	42.46^{CDE}	12.46^{C}

$^{A, B, C, \ldots, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.2 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental legume silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
		Molasses	21.86 ^{F}	87.38 ^{A}	12.59 ^{DEFG}	4.40 ^{BCD}	41.30 ^{EFGH}	39.03^{DEF}	12.62 ^{C}
		FJLB	19.10 ^{F}	87.96 ^{A}	10.99 ^{EFGHIJ}	3.79 ^{EFGHI}	54.36 ^{AB}	49.71^{AB}	12.04 ^{C}
Verano Stylo	30	Control	34.19 ^{AB}	82.21 ^{ABC}	11.53 ^{EFGHI}	3.37 ^{IJKL}	45.63 ^{CDEFG}	26.96^{IJK}	17.79 ^{ABC}
		Cassava meal	36.57 ^{AB}	84.56 ^{ABC}	12.65 ^{DEFG}	3.98 ^{DEFGH}	43.86 ^{DEFGH}	23.51^{KL}	15.45 ^{ABC}
		Molasses	30.02 ^{BCDE}	85.64 ^{AB}	10.98 ^{EFGHIJ}	3.67 ^{FGHIJ}	42.15 ^{DEFGH}	23.10^{KL}	14.36 ^{BC}
		FJLB	32.62 ^{ABC}	77.51 ^{ABC}	10.11 ^{FGHIJ}	3.65 ^{GHIJ}	44.91 ^{CDEFGH}	27.20^{IJK}	22.49 ^{ABC}
	45	Control	31.26 ^{ABCD}	83.32 ^{ABC}	19.03 ^{B}	3.42 ^{IJKL}	47.81 ^{ABCDEF}	34.21^{FGH}	16.68 ^{ABC}
		Cassava meal	33.60 ^{ABC}	75.45 ^{BC}	22.93 ^{A}	2.22 ^{O}	46.79 ^{BCDEF}	34.70^{FGH}	24.55 ^{AB}
		Molasses	32.76 ^{ABC}	82.83 ^{ABC}	16.19 ^{C}	2.66 ^{N}	47.47 ^{ABCDEF}	33.99^{FGH}	17.17 ^{ABC}
		FJLB	31.31 ^{ABCD}	84.91 ^{AB}	15.92 ^{C}	3.06 ^{LMN}	46.28 ^{BCDEF}	37.01^{EFG}	15.09 ^{BC}

$^{A, B, C,\ldots, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.2 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental legume silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
	60	Control	30.30 ^{ABCDE}	84.27 ^{ABC}	13.05 ^{DE}	3.42 ^{IJKL}	49.61 ^{ABCDE}	45.86 ^{ABC}	15.73 ^{ABC}
		Cassava meal	32.80 ^{ABC}	82.54 ^{ABC}	12.08 ^{DEFGH}	3.12 ^{KLMN}	45.56 ^{CDEFG}	43.81 ^{BCD}	17.46 ^{ABC}
		Molasses	31.04 ^{ABCD}	82.30 ^{ABC}	12.98 ^{DE}	3.46 ^{IJKL}	45.44 ^{CDEFG}	44.82 ^{ABC}	17.70 ^{ABC}
		FJLB	29.37 ^{BCDE}	82.30 ^{ABC}	11.93 ^{EFGH}	4.04 ^{DEFGH}	50.53 ^{ABCD}	47.96 ^{ABC}	17.70 ^{ABC}
Cavalcade	30	Control	34.20 ^{AB}	85.10 ^{AB}	9.04 ^{IJ}	5.01 ^{A}	43.66 ^{DEFGH}	26.82 ^{IJK}	14.90 ^{BC}
		Cassava meal	35.57 ^{AB}	73.71 ^{C}	9.72 ^{HIJ}	4.09 ^{DEFG}	41.33 ^{EFGH}	25.03 ^{JKL}	26.30 ^{A}
		Molasses	34.44 ^{AB}	84.94 ^{AB}	10.35 ^{EFGHIJ}	4.06 ^{DEFGH}	40.26 ^{FGH}	24.55 ^{JKL}	15.07 ^{BC}
		FJLB	35.05 ^{AB}	79.13 ^{ABC}	8.68 ^{J}	4.15 ^{CDEF}	42.05 ^{DEFGH}	29.91 ^{HIJ}	20.87 ^{ABC}
	45	Control	35.72 ^{AB}	86.17 ^{AB}	12.41 ^{DEFGH}	2.88 ^{MN}	48.12 ^{ABCDEF}	31.49 ^{GHI}	13.83 ^{BC}
		Cassava meal	37.75 ^{A}	85.11 ^{AB}	12.34 ^{DEFGH}	4.18 ^{CDE}	45.79 ^{CDEFG}	30.15 ^{HIJ}	14.89 ^{BC}

$^{A, B, C,\ldots,.P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.2 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental legume silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
		Molasses	35.61 ^{AB}	83.83 ^{ABC}	12.85 ^{DEF}	4.29 ^{BCDE}	40.48 ^{FGH}	28.96 ^{HIJK}	16.17 ^{ABC}
		FJLB	34.54 ^{AB}	83.42 ^{ABC}	13.03 ^{DE}	4.72 ^{AB}	45.89 ^{CDEFG}	32.19 ^{GHI}	16.59 ^{ABC}
	60	Control	-	-	-	-	-	-	-
		Cassava meal	-	-	-	-	-	-	-
		Molasses	-	-	-	-	-	-	-
		FJLB	-	-	-	-	-	-	-
S.E.M			0.318	0.391	0.095	0.015	0.293	0.211	0.394
S			***	**	***	***	*	***	*
D			**	ns	***	***	***	***	ns
A			ns	ns	ns	ns	***	***	ns

$^{A, B, C,\ldots, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.2 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on chemical compositions of the experimental legume silages.

S	D	A	DM, %	OM, %	CP, %	EE, %	NDF, %	ADF, %	Ash, %
S × D			ns	ns	***	***	ns	**	ns
S × A			ns	ns	***	***	ns	ns	ns
D × A			ns	ns	ns	*	ns	ns	ns
S × D × A			ns	ns	**	***	ns	ns	ns

[A, B, C,…., P]Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.3 Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental grass silages.

S	D	A	pH	NH3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
Purple guineagrass	30	Control	$4.15^{ABCDEFG}$	61.78^{AB}	31.44^{FGHIJ}	5.63^{KLMNO}	1.51^{JK}	1.15^{G}	38.86^{NOPQR}
		Cassava meal	4.27^{ABCDEF}	64.97^{A}	19.58^{JKL}	3.88^{MNO}	1.09^{JK}	0.94^{G}	25.11^{RS}
		Molasses	4.05^{BCDFG}	57.92^{ABC}	80.23^{BC}	6.66^{KLMN}	1.19^{JK}	1.23^{G}	88.56^{DEF}
		FJLB	4.48^{A}	36.80^{EFGHIJ}	45.46^{DEF}	6.10^{KLMNO}	1.08^{JK}	1.14^{G}	52.80^{JKLMN}
	45	Control	$4.18^{ABCDEFG}$	33.53^{FGHIJ}	36.67^{EFGHI}	7.52^{JKLM}	6.59^{FG}	0.00^{G}	50.78^{KLMNO}
		Cassava meal	4.07^{BCDEFG}	32.08^{FGHIJ}	85.71^{B}	13.11^{DEFG}	5.39^{GH}	0.00^{G}	104.61^{ABCD}
		Molasses	3.73^{G}	25.85^{HIJ}	48.22^{DE}	12.21^{EFGH}	5.30^{GH}	0.53^{G}	66.40^{HIJK}
		FJLB	4.41^{ABCD}	32.94^{FGHIJ}	46.93^{DE}	5.33^{LMNO}	5.34^{GH}	0.00^{G}	57.11^{JKLMN}
	60	Control	4.36^{ABCDE}	29.69^{GHIJ}	39.29^{DEFGH}	25.39^{A}	23.94^{A}	4.37^{F}	91.52^{BCDEF}
		Cassava meal	3.94^{DEFG}	22.04^{J}	21.84^{IJKL}	14.52^{CDEF}	13.02^{D}	7.11^{E}	56.49^{JKLMN}

$^{A, B, C,\ldots, R, S}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.3 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental grass silages.

S	D	A	pH	NH3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
		Molasses	$4.16^{ABCDEFG}$	35.59^{FGHIJ}	84.37^{B}	9.22^{HIJK}	9.22^{E}	4.77^{F}	107.56^{ABC}
		FJLB	4.51^{AB}	29.88^{GHIJ}	26.92^{HIJ}	23.94^{A}	25.39^{A}	4.28^{F}	81.96^{FGH}
Mulato II grass	30	Control	4.28^{ABCDEF}	53.45^{ABCDE}	18.89^{JKL}	6.53^{KLMN}	0.58^{K}	1.03^{G}	28.22^{QRS}
		Cassava meal	3.93^{EFG}	67.74^{A}	24.65^{HIJK}	5.37^{LMNO}	0.46^{K}	1.31^{G}	31.41^{PQRS}
		Molasses	4.24^{ABCDEF}	37.95^{EFGHIJ}	80.53^{BC}	4.50^{LMNO}	0.85^{K}	1.30^{G}	85.38^{EFG}
		FJLB	4.29^{ABCDE}	43.44^{CDEFGH}	8.743^{L}	7.72^{JKL}	0.40^{K}	0.98^{G}	17.30^{S}
	45	Control	4.22^{ABCDEF}	46.33^{BCDEFG}	48.34^{DE}	15.67^{CDE}	15.67^{C}	0.00^{G}	84.52^{EFG}
		Cassava meal	4.01^{DEFG}	36.50^{FGHIJ}	81.12^{BC}	10.66^{GHIJ}	16.56^{C}	0.00^{G}	108.34^{AB}
		Molasses	3.30^{H}	35.57^{FGHIJ}	102.03^{A}	7.76^{JKL}	8.32^{E}	0.00^{G}	118.11^{A}
		FJLB	4.28^{ABCDEF}	48.28^{BCDEF}	52.36^{D}	24.53^{A}	24.53^{A}	0.00^{G}	101.42^{ABCDE}

$^{A, B, C,\ldots, R, S}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.3 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental grass silages.

S	D	A	pH	NH3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
Mulato II grass	60	Control	4.49^{ABC}	21.01^{J}	25.18^{HIJK}	11.30^{FGHI}	4.43^{HI}	0.00^{G}	41.29^{MNOPQR}
		Cassava meal	4.02^{BCDEFG}	33.31^{FGHIJ}	48.17^{DE}	5.69^{KLMNO}	6.18^{GH}	0.23^{G}	59.79^{IJKLM}
		Molasses	$4.13^{ABCDEFG}$	$42.25^{CDEFGHI}$	39.74^{DEFGH}	16.98^{BC}	8.57^{E}	3.84^{F}	69.87^{GHIJ}
		FJLB	4.41^{ABCD}	40.60^{DEFGHI}	31.30^{FGHIJ}	14.23^{CDEF}	6.14^{GH}	0.00^{G}	50.71^{KLMNO}
Napiergrass	30	Control	$4.17^{ABCDEFG}$	27.34^{HIJ}	26.68^{HIJ}	2.48^{O}	1.11^{JK}	25.20^{A}	54.02^{JKLMN}
		Cassava meal	4.11^{BCDEFG}	58.31^{ABC}	45.03^{DEF}	3.33^{NO}	0.86^{K}	10.80^{CD}	60.32^{IJKL}
		Molasses	4.02^{DEFG}	55.57^{ABCD}	68.84^{C}	3.70^{NO}	1.60^{JK}	12.50^{BC}	90.05^{CDEF}
		FJLB	4.10^{BCDEFG}	25.33^{IJ}	30.10^{GHIJ}	2.98^{NO}	0.91^{K}	10.36^{D}	44.28^{LMNOPQ}
	45	Control	3.83^{FG}	33.10^{FGHIJ}	43.58^{DEFG}	12.03^{FGH}	12.54^{D}	7.30^{E}	75.86^{FGHI}

$^{A, B, C,\ldots, R, S}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.3 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental grass silages.

S	D	A	pH	NH3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
		Cassava meal	3.74^{G}	36.01^{FGHIJ}	10.59^{KL}	16.36^{CD}	16.35^{C}	4.40^{F}	47.68^{LMNOP}
		Molasses	3.33^{H}	20.75^{J}	75.44^{BC}	3.11^{NO}	3.11^{IJ}	4.11^{F}	85.77^{EFG}
		FJLB	4.04^{CDEFG}	31.49^{FGHIJ}	47.90^{DE}	19.81^{B}	19.81^{B}	0.00^{G}	87.52^{DEFG}
	60	Control	4.04^{CDEFG}	31.95^{FGHIJ}	8.155^{L}	5.50^{LMNO}	5.13^{GH}	14.16^{B}	33.34^{OPQRS}
		Cassava meal	3.95^{DEFG}	28.75^{GHIJ}	29.92^{GHIJ}	5.13^{LMNO}	1.49^{JK}	9.49^{D}	46.23^{LMNOPQ}
Napiergrass	60	Molasses	3.97^{DEFG}	26.50^{HIJ}	31.94^{FGHIJ}	8.10^{IJKL}	0.20^{K}	5.42^{EF}	45.66^{LMNOPQ}
		FJLB	3.96^{DEFG}	32.11^{FGHIJ}	23.46^{IJK}	4.56^{LMN0}	1.33^{JK}	4.34^{F}	33.53^{OPQRS}
S.E.M			0.022	1.011	1.761	0.332	0.105	0.102	0.922
S			***	**	***	***	***	***	***
D			***	***	***	***	***	***	***

$^{A, B, C,\ldots, R, S}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.3 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental grass silages.

S	D	A	pH	NH3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
A			***	ns	***	***	***	***	***
S × D			ns	*	**	***	***	***	***
S × A			ns	ns	***	***	***	***	*
D × A			**	***	***	***	***	***	***
S × D × A			ns	**	***	***	***	***	***

[A, B, C,....., R, S]Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant different (P>0.05) and SEM: standard error of mean.

Table 3.2.4 Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental legume silages.

S	D	A	pH	NH_3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
Thapra Stylo	30	Control	$4.66^{ABCDEFGH}$	20.56^{BC}	$35.27^{FGHIJKL}$	6.84^{IJK}	1.15^{HIJ}	1.51^{HI}	39.00^{MNO}
		Cassava meal	$4.48^{DEFGHIJK}$	33.95^{AB}	$33.24^{FGHIJKL}$	12.63^{F}	1.14^{HIJ}	1.09^{I}	$59.48^{GHIJKLM}$
		Molasses	4.14^{IJK}	26.05^{BC}	63.14^{BCD}	13.09^{FE}	1.23^{HIJ}	1.19^{HI}	76.89^{EFG}
		FJLB	4.97^{ABCDE}	28.89^{CB}	45.67^{DEFGH}	11.67^{FG}	1.14^{HIJ}	1.32^{HI}	60.33^{GHIJKL}
	45	Control	4.99^{ABCD}	43.49^{A}	41.78^{EFGHIJ}	8.79^{GHI}	8.79^{G}	11.24^{EF}	74.50^{EFGH}
		Cassava meal	$4.60^{BCDEFGHIJK}$	44.01^{A}	44.19^{EFGHI}	27.38^{B}	27.38^{B}	6.84^{FGH}	105.15^{BCD}
		Molasses	4.93^{ABCDEF}	20.48^{BC}	54.22^{CDE}	11.45^{FG}	11.45^{EF}	7.24^{FG}	83.92^{EF}
		FJLB	$4.73^{ABCDEFGH}$	21.51^{BC}	51.72^{CDEF}	10.54^{FGH}	10.54^{FG}	3.13^{GHI}	67.84^{FGHIJ}
	60	Control	5.21^{A}	25.89^{BC}	7.07^{N}	6.33^{IJKL}	2.60^{HI}	76.03^{A}	91.02^{CDE}
		Cassava meal	4.86^{ABCDEF}	25.24^{CB}	48.76^{CDEFG}	7.07^{IJ}	3.17^{H}	13.66^{E}	72.65^{EFGHI}

$^{A, B, C,\ldots., O, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested.

Table 3.2.4 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental legume silages.

S	D	A	pH	NH_3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
		Molasses	4.35 ^{FGHIJK}	21.58^{BC}	22.32^{KLMN}	23.28^{C}	0.79^{IJ}	0.00^{I}	46.79^{KLMNO}
		FJLB	5.11 ^{AB}	25.84^{BC}	13.27^{MN}	3.99^{JKL}	0.00^{J}	0.00^{I}	17.26^{P}
Verano Stylo	30	Control	4.63 ^{BCDEFGHIJ}	44.32^{A}	27.19^{HIJKLM}	5.16^{JKL}	1.03^{HIJ}	0.69^{I}	35.00^{NOP}
		Cassava meal	4.30 ^{GHIJK}	25.09^{BC}	32.84^{GHIJKL}	3.47^{KLM}	1.31^{HIJ}	0.67^{I}	36.74^{NOP}
		Molasses	4.04 ^{K}	21.90^{BC}	41.58^{EFGHIJ}	2.98^{LMN}	1.30^{HIJ}	0.77^{I}	43.02^{LMNO}
		FJLB	4.62 ^{BCDEFGHIJ}	24.22^{BC}	40.17^{EFGHIJK}	11.87^{FG}	0.98^{IJ}	0.83^{I}	53.36^{IJKLMNO}
	45	Control	5.06 ^{ABC}	28.42^{BC}	34.33^{FGHIJKL}	31.22^{A}	31.22^{A}	55.75^{B}	143.09^{A}
		Cassava meal	4.49 ^{DEFGHIJK}	25.34^{BC}	41.08^{EFGHIJ}	18.51^{D}	12.74^{E}	11.36^{EF}	83.69^{EF}
		Molasses	4.81 ^{ABCDEFG}	26.57^{BC}	48.89^{CDEFG}	16.06^{DE}	12.46^{EF}	0.00^{I}	75.67^{EFGH}
		FJLB	4.46 ^{DEFGHIJK}	21.46^{BC}	48.04^{CDEFG}	23.68^{C}	23.48^{C}	10.99^{EF}	106.18^{BCD}

$^{A, B, C,\ldots., O, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested.

Table 3.2.4 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental legume silages.

S	D	A	pH	NH_3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
Verano Stylo	60	Control	5.03^{ABCD}	21.67^{CB}	13.93^{MN}	7.00^{IJ}	0.00^{J}	10.85^{EF}	32.88^{OP}
		Cassava meal	$4.62^{BCDEFGHIJ}$	20.29^{BC}	21.41^{LMN}	7.52^{HIJ}	0.00^{J}	12.44^{EF}	41.36^{LMNO}
		Molasses	4.15^{HIJK}	28.02^{BC}	24.86^{JKLM}	11.08^{FG}	1.90^{HIJ}	15.79^{E}	$55.57^{HIJKLMN}$
		FJLB	$4.65^{ABCDEFGHIJ}$	26.29^{BC}	26.35^{IJKLM}	6.49^{IJK}	0.00^{J}	13.74^{E}	47.49^{JKLMNO}
Cavalcade	30	Control	$4.62^{BCDEFGHIJ}$	27.01^{BC}	50.08^{CDEFG}	7.09^{IJ}	25.20^{C}	1.19^{HI}	86.49^{DEF}
		Cassava meal	$4.51^{DEFGHIJK}$	27.38^{BC}	50.52^{CDEFG}	4.58^{JKL}	10.80^{EF}	0.86^{I}	65.20^{FGHIJK}
		Molasses	4.07^{JK}	25.67^{BC}	72.14^{B}	4.80^{JKL}	15.57^{D}	1.60^{HI}	90.69^{CDE}
		FJLB	$4.51^{CDEFGHIJK}$	26.41^{BC}	64.17^{BC}	5.11^{JKL}	14.77^{D}	0.91^{I}	86.25^{DEF}
	45	Control	$4.38^{EFGHIJK}$	29.06^{BC}	$35.27^{FGHIJKL}$	0.10^{N}	0.23^{J}	41.79^{C}	77.38^{EFG}
		Cassava meal	$4.78^{ABCDEFG}$	23.21^{BC}	88.94^{A}	0.30^{MN}	0.00^{J}	16.11^{E}	105.35^{BCD}

$^{A, B, C,\ldots, O, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested.

Table 3.2.4 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental legume silages.

S	D	A	pH	NH_3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
		Molasses	4.87^{ABCDEF}	22.48^{BC}	92.31^{A}	0.00^{N}	0.00^{J}	25.79^{D}	118.10^{B}
		FJLB	4.34^{GHIJK}	18.66^{C}	93.45^{A}	0.36^{MN}	0.00^{J}	14.59^{E}	108.39^{BC}
Cavalcade	60	Control	-	-	-	-	-	-	-
		Cassava meal	-	-	-	-	-	-	-
		Molasses	-	-	-	-	-	-	-
		FJLB	-	-	-	-	-	-	-
S.E.M			0.028	0.872	0.858	0.269	0.109	0.295	1.118
S			ns	ns	***	***	***	***	***
D			ns	ns	***	***	***	***	***
A			***	**	***	*	***	***	ns

$^{A, B, C,\ldots, O, P}$Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested.

Table 3.2.4 (Cont.) Effect of species (S), cutting date (D, days after regrowth) and additive (A) on fermentation quality of the experimental legume silages.

S	D	A	pH	NH_3-N, g/kg DM	Lactic acid, g/kg DM	Acetic acid, g/kg DM	Propionic acid, g/kg DM	Butyric acid, g/kg DM	Total VFA, g/kg DM
S × D			*	ns	*	***	***	***	***
S × A			ns	ns	**	***	***	***	***
D × A			ns	*	***	***	***	***	***
S × D × A			ns	*	**	***	***	***	***

[A, B, C,…., O, P]Means followed by a different letter within the same column are significantly different: *P<0.05; **P<0.01; ***P<0.001, ns: not significant difference (P>0.05), SEM: standard error of mean and - = can not be harvested.

Table 3.2.5 Effect of species (S) and additives (A) on fatty acid composition (g/100 g total fat) in the experiment grass silage.

	Purple guineagrass				Mulato II grass				Napiergrass				SEM	S	A	S × A
	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB				
C12:0	0.56^{BC}	0.68^{B}	0.41^{CD}	0.01^{F}	0.94^{A}	0.17^{EF}	0.41^{CD}	0.25^{DE}	0.23^{DE}	0.01^{F}	0.46^{C}	0.04^{F}	0.012	***	***	***
C14:0	0.81^{A}	0.99^{A}	1.52^{A}	0.74^{A}	1.36^{A}	1.56^{A}	1.07^{A}	0.78^{A}	0.56^{A}	1.30^{A}	0.75^{A}	0.57^{A}	0.099	ns	ns	ns
C15:0	0.05^{D}	0.18^{CD}	0.05^{D}	0.08^{D}	0.01^{D}	0.27^{CD}	0.30^{CD}	1.01^{B}	0.41^{C}	0.00^{D}	0.45^{C}	1.71^{A}	0.020	***	***	***
C16:0	15.62^{C}	18.58^{ABC}	16.41^{BC}	18.83^{ABC}	20.71^{ABC}	17.35^{ABC}	20.45^{ABC}	22.86^{A}	19.03^{ABC}	17.74^{ABC}	21.88^{AB}	18.15^{ABC}	0.459	*	ns	ns
C16:1	1.45^{A}	1.44^{A}	1.21^{AB}	0.13^{C}	1.48^{A}	0.46^{C}	0.68^{BC}	1.45^{A}	0.15^{C}	1.61^{A}	1.53^{A}	1.39^{A}	0.061	ns	ns	***
C17:0	0.29^{A}	0.16^{AB}	0.02^{B}	0.03^{B}	0.25^{A}	0.16^{AB}	0.23^{A}	0.23^{A}	0.19^{A}	0.02^{B}	0.31^{A}	0.02^{B}	0.013	*	***	***
C18:0	2.24^{A}	1.80^{A}	1.37^{AB}	0.54^{B}	1.32^{AB}	1.81^{A}	1.84^{A}	2.09^{A}	1.55^{AB}	1.36^{AB}	1.58^{AB}	2.23^{A}	0.099	ns	ns	**
C18:1n9	1.82^{B}	2.00^{B}	1.81^{B}	1.84^{B}	1.67^{B}	1.69^{B}	2.31^{B}	2.35^{B}	1.87^{B}	2.34^{B}	3.97^{A}	2.54^{B}	0.098	***	*	ns
C18:2n6	15.22^{ABCD}	13.73^{CD}	16.80^{AB}	17.17^{A}	13.67^{ABCD}	14.89^{CD}	15.92^{ABC}	14.49^{BCD}	15.17^{ABCD}	13.12^{D}	15.78^{ABC}	17.23^{A}	0.220	ns	*	ns
C18:3n3	49.98^{A}	51.42^{A}	49.97^{A}	43.63^{AB}	45.39^{AB}	49.80^{A}	45.94^{AB}	41.57^{B}	49.47^{AB}	48.82^{AB}	45.52^{AB}	44.67^{AB}	0.683	ns	*	ns

$^{A, B, C, D, E, F}$ Means followed by a different letter within the same row are significantly different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

Table 3.2.5 (Cont.) Effect of species (S) and additives (A) on fatty acid composition (g/100 g total fat) in the experiment grass silage.

	Purple guineagrass				Mulato II grass				Napiergrass				SEM	S	A	S × A
	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB				
SFA	19.80^{B}	21.97^{AB}	20.83^{B}	20.27^{B}	23.69^{AB}	21.86^{AB}	24.45^{AB}	27.12^{A}	21.97^{AB}	19.75^{B}	25.26^{AB}	22.70^{AB}	0.484	*	ns	ns
MUFA	3.10^{BC}	3.45^{B}	3.02^{BC}	1.97^{C}	3.15^{BC}	2.15^{C}	2.99^{BC}	3.80^{B}	2.01^{C}	3.95^{B}	5.51^{A}	3.92^{B}	0.104	**	**	***
PUFA	65.19^{A}	65.15^{A}	66.77^{A}	60.80^{AB}	59.07^{AB}	65.36^{A}	61.85^{AB}	56.06^{B}	64.63^{A}	61.95^{AB}	61.30^{AB}	61.90^{AB}	0.686	*	ns	ns
PUFA/SFA	3.06^{A}	3.03^{A}	2.94^{A}	3.09^{A}	2.54^{A}	3.28^{A}	2.63^{A}	2.07^{A}	2.95^{A}	3.14^{A}	2.59^{A}	2.73^{A}	0.082	ns	ns	ns

$^{A, B, C, D, E, F}$ Means followed by a different letter within the same row are significant different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3

Table 3.2.6 Effect of species (S) and additives (A) on fatty acid composition (g/100 g total fat) in the experiment legume silage.

	Thapra stylo				Verano stylo				Cavalcade				SEM	S	A	S × A
	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB				
C12:0	0.24^{AB}	0.30^{A}	0.08^{BC}	0.33^{A}	0.05^{C}	0.25^{A}	0.02^{C}	0.18^{ABC}	0.024^{C}	0.02^{C}	0.03^{C}	0.03^{C}	0.100	***	**	ns
C14:0	0.72^{B}	0.69^{BC}	0.57^{BC}	0.55^{BC}	0.55^{BC}	0.53^{BC}	0.60^{BC}	0.39^{C}	1.41^{A}	0.69^{BC}	0.53^{BC}	1.45^{A}	0.152	***	**	***
C15:0	0.42^{A}	0.25^{BC}	0.04^{D}	0.12^{CD}	0.04^{D}	0.33^{AB}	0.21^{BCD}	0.06^{D}	0.06^{D}	0.09^{CD}	0.06^{D}	0.06^{D}	0.015	**	**	***
C16:0	19.74^{AB}	20.11^{AB}	18.40^{AB}	15.86^{B}	20.05^{AB}	22.77^{A}	18.35^{AB}	18.70^{AB}	14.97^{B}	16.9^{B}	16.28^{B}	16.37^{B}	0.485	**	ns	ns
C16:1	1.70^{A}	0.56^{B}	0.18^{B}	1.69^{A}	1.25^{A}	0.33^{B}	0.48^{B}	1.27^{A}	1.53^{A}	1.36^{A}	0.39^{B}	1.35^{A}	0.060	ns	***	ns
C17:0	0.26^{ABCDE}	0.24^{BCDE}	0.06^{E}	0.39^{ABC}	0.04^{E}	0.35^{ABCD}	0.03^{E}	0.18^{CDE}	0.47^{AB}	0.15^{DE}	0.50^{A}	0.32^{ABCD}	0.021	**	ns	***
C18:0	4.18^{ABC}	3.37^{BCD}	1.50^{E}	3.56^{BCD}	2.65^{CDE}	2.31^{DE}	2.95^{CDE}	2.69^{CDE}	4.82^{AB}	4.69^{AB}	5.42^{A}	5.11^{AB}	0.150	***	ns	ns
C18:1n9	3.31^{AB}	4.40^{A}	3.75^{AB}	3.24^{AB}	3.38^{AB}	3.50^{AB}	1.27^{B}	3.47^{AB}	3.98^{A}	3.17^{AB}	4.66^{A}	2.38^{AB}	0.210	ns	ns	*
C18:2n6	17.57^{B}	16.38^{B}	18.75^{B}	17.14^{B}	24.29^{A}	21.25^{AB}	25.98^{A}	21.25^{AB}	18.27^{B}	16.99^{B}	17.34^{B}	18.19^{B}	0.482	***	ns	ns
C18:3n3	37.50^{AB}	37.23^{AB}	45.02^{A}	36.65^{AB}	35.00^{B}	38.40^{AB}	41.52^{AB}	44.90^{A}	43.69^{A}	43.04^{AB}	43.30^{AB}	42.77^{AB}	0.691	*	ns	ns

$^{A, B, C, D, E}$ Means followed by a different letter within the same row are significantly different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3

Table 3.2.6 (Cont.) Effect of species (S) and additives (A) on fatty acid composition (g/100 g total fat) in the experiment legume silage.

	Thapra stylo				Verano stylo				Cavalcade				SEM	S	A	S × A
	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB	Control	Cassava	Molasses	FJLB				
SFA	25.40 ^{A}	24.83 ^{A}	20.62 ^{A}	20.66 ^{A}	23.46 ^{A}	26.41 ^{A}	22.27 ^{A}	22.07 ^{A}	21.76 ^{A}	22.58 ^{A}	22.85 ^{A}	23.38 ^{A}	0.515	ns	ns	ns
MUFA	5.02 ^{A}	4.97 ^{A}	3.93 ^{A}	4.92 ^{A}	5.30 ^{A}	3.97 ^{A}	1.75 ^{B}	4.71 ^{A}	5.51 ^{A}	4.53 ^{A}	4.39 ^{A}	3.72 ^{AB}	0.195	ns	*	ns
PUFA	55.07 ^{BC}	53.61 ^{C}	63.77 ^{AB}	53.78 ^{C}	59.30 ^{ABC}	59.65 ^{ABC}	67.50 ^{A}	66.16 ^{A}	61.96 ^{ABC}	60.03 ^{ABC}	60.63 ^{ABC}	60.96 ^{ABC}	0.766	**	*	ns
PUFA/SFA	2.18 ^{B}	2.16 ^{B}	3.11 ^{A}	2.66 ^{AB}	2.39 ^{AB}	2.60 ^{AB}	3.06 ^{A}	3.05 ^{A}	2.88 ^{AB}	2.66 ^{AB}	2.65 ^{AB}	2.62 ^{AB}	0.065	ns	*	ns

$^{A, B, C, D, E}$ Means followed by a different letter within the same row are significantly different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean, SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3

3.2.6 Discussion

Species of grass in the current study had influences on all chemical composition, except for the ADF of the grass silage. The Purple guineagrass silage contains high contents of DM, OM and NDF, while the Mutato II grass silage contained high DM, CP, EE and ash. However, the Napiergras silage had low DM and NDF and high in CP and ash. When fermentation quality of final grass silages was considered, there were higher pH, NH_3-N, LA, AA, PA and total VFAs but lower BA for the Purple guineagrass silage and the Mulato II grass silage, when compared with those of the Napiergrass silage. The DM content in the grass silage was ranged from 16.64-28.64%, while the DM content of fresh grasses was 20.35-31.14% (data from the Experiment I-I), indicating a little losses of DM during grass ensiling. The chemical composition of the grass silages with advanced maturity seemed to be related to the chemical contents in fresh grasses as reported in the Experiment I-I; lowering CP and EE but increasing DM, NDF, ADF and ash with advancing maturity. However, there were inconsistent with the days of regrowth. This might be explained by the additive added into the fresh grasses for ensiling and fermentative processes during ensiling. There were low pH, NH_3-N and BA, but high in LA, AA, PA and total VFAs for the grass harvested at 45 days after regrowth, indicating proper age of regrowth for giving fermentation quality of the grass ensiling. The molasses added for ensiling had clearly lowering NDF content of grass silage while there were inconsistent effects on chemical composition for the other additives. Adding molasses for grass ensiling also obviously increased LA and then total VFAs, while the FJLB supplementation increased AA and PA proportions but lowered BA contents. These would be explained by the fact that molasses contained large proportion of sugar (Olbrich, 1963, and

Bureenok, et al., 2005b) used as substrate for LA formation (Alli etl al., 1985) and the grasses had LAB (Bureenok et al., 2005a, and Tao et al., 2012), resulting in propagation of LA bacteria to inhibit the growth of clostridia and aerobic bacteria (Wang et at., 2009).

Well preserved silage types could be characterized as low pH (<5), low ammonia N (<90 g/kg of total N) and low concentration of butyric acid (<5.5 g/kg DM) as concluded by Phiri et al. (2007). On overview, grass silages in the present study had the ranges of pH 3.30-4.51, NH_3-N 20.75-67.74 g/kg of total N and butyric acid 0.00-25.20 g/kg DM. This would indicated that the grass silages at all days of regrowth and with all additive supplementation had pH and NH_3-N in the normal range of the criteria for well preserved silage of grasses, but the butyric acid concentration in some grass silages was above 5.5 g/kg DM, which implied losses of some nutrients and shorter storage duration of these grass silages. From the Table 3.2.3, there was quite high for the content of the total VFAs in the Mulato II grass silage at the 45 days after regrowth with no detectable butyric acid and high proportion of LA, AA and PA. This would be reasonable to choose the Mulato II grass silage prepared from the 45 days after regrowth. In the Experiment I-I, there was no difference for chemical composition of fresh grass, except for rather high CP content in Mulato II grass harvested at 45 days after regrowth. Therefore, the change of chemical composition and fermentation quality during ensiling would be mainly caused by the additive supplementation. The cassava meal contained mainly starch while the molasses had high proportion of sugar. Both starch and sugar are the important substrates for LA formation in fermentation during ensiling, especially at the initial stage of anaerobic fermentation (Driehuis and Oude Elferink, 2000). These

lead to quite high proportion of lactic acid (81.12 and 102.03 g/kg DM, respectively) in the silage of Mulato II grass harvested at 45 days after regrowth. For the FJLB additive, the main composition is LAB with a small proportion of glucose added in the solution of the FJLB. Consequently, the less substrate providing for fermentation resulted in no difference of the LA formation when compared with the Mulato II grass silage without additive (the Control). However, there was highest proportion of AA formation in the Mulato II grass silage at 45 days after regrowth. The high formation of AA content might be the results of heterofermentive LAB in the FJLB taking place for producing more acetic acids (McDonalds et al., 1991; Kaiser et al., 2004; Driehuis and Oude Elferink, 2000). For PA formation, it is possible that propionic acid bacteria can ferment sugars and lactate to acetate and propionate (Higginbotham et al., 1998). These short-chain aliphatic acids inhibit the growth of yeasts and molds in silage (Woolford, 1975; Moon, 1983).

For legume silage, the legume species had influence on all analyzes chemical compositions. The DM content in the legume silage was ranged from 19.10-37.75%, while the DM content of fresh grasses was 22.15-32.56% (data from the Experiment I-I), implying not many losses and a little gains of DM during legume ensiling. The Thapra stylo silage contained rather low DM content but high in concentration of CP, NDF and ADF. The Verano stylo and Cavalcade silages had high DM and ash content. The the Verano stylo silage contained high CP, NDF and ADF, while these contents were low for the Cavalcade silage. When the fermentation quality of final legume silages was considered, there was an effect of legume species on formation of VFAs. The Cavalcade silage contained higher proportion of LA and total VFAs than the silages of the Thapra stylo and Verano stylo. In the meantime, the Cavalcade silage

had lower AA content than the Thapra stylo and Verano stylo silages. The chemical composition of the legume silages with advanced stage of maturity was seemed to be related to the chemical profiles for NDF and ADF in fresh legumes as reported in the Experiment I-I, while the final legume silages with advancing maturity had a trend of higher CP and EE; especially the CP contents at 45 days after regrowth and EE contents at 45 and 60 days after regrowth. However, the total CP contents in legume silage decreased; indicating nitrogen content in fresh legumes reduced during fermentation processes. The individual and total VFAs of the legume silages of 45 days after regrowth were higher than those of 30 and 60 days after regrowth, except for the rather high BA content for both 45 and 60 days after regrowth. These results were different from the fermentation quality of the studied grass silages as higher values of pH and proportion of BA. These would be explained by the fact that high buffering capacity (McDonald and Henderson, 1962) and low concentration in water soluble carbohydrates (Dewhurst et al., 2003) in legumes with the risk of a BA fermentation (Hattori et al., 1996). There were rather evident that the legumes supplemented with each additive improved fermentation quality compared with the legumes without additive supplementation as decreasing pH value, NH_3-N and BA whereas increasing LA and AA.

Comparing with well preserved silage types as mentioned earlier (pH<5), NH_3-N (<90 g/kg of total N) and concentration of BA (<5.5 g/kg DM), the legume silages had overall ranges of pH 4.04-5.21, NH_3-N 18.66-44.32 g/kg of total N and BA 0.00-76.03 g/kg DM. The production of NH_3-N values of legume silages was rather low, compared to those of the grass silages, attributing to the high proportion of total N resulted in low proportion of NH_3-N/total N This would indicated that the legume

silages at some cutting date and/or some silage additives, including interaction of studied factors, did not meet the criteria (pH and butyric acid) for well preserved silages. The silages of the legumes harvested at 45 days after regrowth had high concentration of LA with acceptable high pH, low NH_3-N and higher BA content compared with the criteria. At 45 days after regrowth, the Cavalcade silage had the highest values of LA concentration, while the Thapra stylo and the Verano stylo silages had LA at the level of close to each other. However, the Cavalcade silage at 45 days after regrowth had rather high BA concentration with very low AA concentration, thus there are risk for protein degradation and spoilage from *Clostridium* spp., molds and yeasts (Kaiser et al., 2004). The NH_3-N production of Thapra stylo silage at 45 days after regrowth was higher than the Verano stylo and the Cavalcade silages at the same cutting date. When the additive effect was considered for the silage of Verano stylo harvested at 45 days after regrowth, the FJLB had highest LA formation and lowest NH_3-N content, with acceptable pH value. Therefore, from overall properties, the Verano stylo harvested at 45 days after regrowth with the FJLB additive would be chosen for ensiling to preserve legume forage for the ruminants. However, the rather high pH and BA would lead to awareness for risk of low activity of bacteria and spoilage of silages.

From the current study, the Mulato II grass and the Verano stylo, harvested at 45 days after regrowth were suitable forage for making silage for ruminants as their chemical composition and fermentation quality of the final silages. For experimental additives, the molasses and cassava meal were effective additives but their high cost and difficulty for preparing would lead to undesirable additive for formers. Adding FJLB into Mulato II grass and Verano stylo would benefit for storing silage during

supply for ruminants, although not much increase of LA contents. The FJLB would be an alternative additive as the low cost and easy preparing for farmers.

Concentration of PUFA in grass and legume silages was predominant. The C18:3n3 was the main proportion of PUFA in both silages. This would be explained by the fact that the membrane glycerolipids of grasses are dominated by C18:3n3 (0.58±0.160 g/g total FAs) as the reports earlier (Chilliard et al., 2001; Van Ranst, 2009). Structure of fresh legumes had rather similarity with the structure of grasses, thus the high proportion of C18:3n3 in legume silages would be the results of the C18:3n3 contents in the membrane glycerolipids. However, there were highly variable content of C18:3n3 with the silage additives. In the current study, the Mulato II grass silage treated with FJLB had the lowest concentration of C18:3n3 among all additives added, while the Verano stylo silage treated with FJLB had the highest of C18:3n3 among all additives added. In the meantime, there was no effect of additives on content of C18:3n3 in the experimental grass and legume silages. These imply all studied additives would maintain content of C18:3n3 in silages of grasses and legumes.

In ruminal fate of lipids, the formation of conjugated fatty acids (CLA) was occurred by bacterial in rumen, mainly originated from C18:2n6 and C18:3n3 (Buccioni et al., 2012; Kishino et al., 2009; Ogawa et al., 2005). In the present study, the contents of C18:2n6 in the grass and legume silages had no systematic pattern with additive treatments. The difference of C18:2n6 concentration was found in the Napiergrass silage treated with FJLB and cassava meal. Hence, the experimental additive would preserve content of C18:2n6. The grass and legume silages treated with additives or without additive are the sources for supplying both C18:2n6 and C18:3n3,

used for formation of CLA in rumen of the ruminants. The other experimental FAs (C16:0, C18:0 and C18:1n9) in the studied grass and legume silages were also no systematic pattern with additive treatments, indicating all experimental additives could be used for ensiling.

On overview, there were small numerical changes of individual FA contents in all experiment treatments. The report of Khan et al. (2012) has shown that bruising, silage pH and NH_3-N content did not affect the content of FA of the grass silages. This would be explained for the results of small numerical changes of FA contents obtained from the current study for both grass and legume silages. The results of the study of Arvidsson et al. (2009) who found no effect of timothy ensiling on key FAs (C16:0, C18:0, C18:1, C18:2 and C18:3), which supported the results of the current study. The lipid composition of herbage are usually associated with cell membranes, called membrane lipids (glycolipids and phospholipids), which the glycolipids bounded with carbohydrate (Buccioni et al., 2012). These would imply that the fermentation process during ensiling had no influences on large change of FA contents composed of cell membranes of forages.

3.2.7 Conclusions

The cutting date at 45 days after regrowth for both Mulato II grass and Verano stylo was suitable for ensiling. The FJLB was not the most effective additive, but low cost and feasible additive for farmers, leading to be an alternative additive.

Grasses (Purple guinea, Mulato II and Napier) and legumes (Thapra stylo, Verano stylo and Cavalcade) harvested at 45 days after regrowth with additive treatments of cassava meal, molasses and FJLB during ensilage had a little effect on

numerical changes of key FA contents (C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3). The studied additives (cassava meal, molasses and FJLB) would preserve FA contents of grass and legume silages.

3.2.8 References

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CHAPTER IV

EXPERIMENT II

EFFECT OF FRESH AND SILAGES OF MULATO II GRASS AND VERANO STYLO ON INTAKE AND RUMEN FERMENTATION OF MEAT

4.1 Abstract

The current study was conducted to study effect of fresh forages and forage silages of Mulato II Grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*) and Verano Stylo (*Stylosanthes hamata*) on intake and rumen fermentation of meat goats. Two forages source for this study were Mulato II grass and Verano stylo and two formations of forage (fresh and silage prepared by adding fermented juice of epiphytic lactic acid bacteria; FJLB). The silage making was prepared and allowed to be fermented for 80 days at room temperature. Eight male ruminally fistulated crossbred Boer × Anglo-Nubian goats (approximately 22.8 kg average body weights) were used as randomly assigned as 2 × 2 factorial arrangements in a 4 × 4 replicated Latin square design to receive four dietary treatments; 1) Fresh Mulato II grass, 2) Fresh Verano stylo, 3) Mulato II grass silage and 4) Verano stylo silage. Each period was lasting for 28 days including the first 7 d used as adjustment period. The results showed that the goats fed on the Mulato II grass silage had highest ($P<0.05$) intake of roughage and total intake. The rumen characteristics were changed with higher time post feeding, especially at the first 2 hours post feeding. There were no effects of dietary treatments

on rumen characteristics, except for a few parameters at particular hours post feeding. The goats fed with the Verano stylo had higher ($P<0.05$) concentration of BUN that those offered the Mulato II grass. There were no differences ($P>0.05$) of fatty acid (FA) profiles of rumen fluid among dietary treatments, but FA profiles in the rumen have been changed by extremely increasing proportion of C18:0 and greatly lowering proportion of C18:2n6 and C18:3n3.

Key Words: Mulato II grass, Verano stylo, Rumen characteristic and microorganism, Fatty acid profiles, Meat goats

4.2 Introduction

The proportion of saturated fatty acid (SFA) in ruminant muscle lipids are often high (Bas et al., 1996) and the polyunsaturated fatty acid (PUFA)/SFA ratio is lower because dietary unsaturated fatty acid (USFA) is hydrogenated in the rumen (Jenkins et al., 2008; Lee et al., 2006). The n-6 and n-3 FA containing in dietary fats are rapidly hydrolyzed by rumen microorganisms and hydrogenated to mainly stearic acid (C18:0). Small amounts of USFA taken up by the microbes will escape hydrogenation in the fore-stomachs, the predominant *trans*-11 C18:1 can be converted into *cis*-9, *trans-11* CLA by the enzyme Δ^9-desaturase in the mammary gland and adipose tissue, and it is thought that this route forms the majority of *cis*-9, *trans*-11 CLA found in ruminant meat and milk (Jakobsen, 1999; Lee et al., 2006; Piperova et al., 2002). Recently, it has been known that increase of dietary PUFA for ruminants in order to escalate PUFA in ruminant meat was limited by microorganisms in rumen. However, numerous studies with ruminants show that feeding forages to ruminants

increases the n-3 PUFA content in milk and meat (Dewhurst et al., 2003; Dewhurst et al., 2006) as they are natural rich sources of C18:3 n3. Nutritional treatments can be used to manipulate the fatty acid (FA) content of muscle to improve the nutritional balance in ruminants with the challenge to increase the PUFA/SFA value (Atti et al., 2006). In tropical area, there is constraint in roughage production during dry season. Silage making is an alternative to preserve quality of roughages for feeding the ruminant. However, fermentation process during ensilage might affect the proportion of chemical and fatty composition in forage silage. Goat is a ruminant, which also be influenced by the rumen microorganisms. The forage sources and ensiling were hypothesized that they could be affected to the production of meat goats and rumen ecology. Thus, the current study was aimed investigate the effect of forage source and ensilage of Mulato II grass and Verano stylo on growth performance and rumen ecology of meat goats.

4.3 Materials and methods

4.3.1 Plant materials

The grass and legume used in the current study were Mulato II grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*) and Verano Stylo (*Stylosanthes hamata*). A series of 10 plots (each 20 m × 20 m) was prepared with surrounding defense and watering system. The 10 plots were allocated into two groups equally; 5 plots for sowing Mulato II grass and the other 5 plots for sowing Verano stylo. There was no fertilizer applied and the forages were sown on June 2010 at Institute of Agricultural Technology, Suranaree University of Technology, Nakhon Ratchasima.

4.3.2 Silage making

After forage harvesting, the experimental forages were immediately chopped into 1-2 cm-lenght pieces. Then fermented juice of epiphytic lactic acid bacteria (FJLB) added at 1% of fresh matter as a silage additive while no additive added for the control grass and legume silages. Approximate 80 kg of grass or legume compressed in plastic bulk. Thirty replicated plastic bulks per each treatment were prepared and allowed to be fermented for 80 days at room temperature.

4.3.3 Fermented juice of epiphytic lactic acid bacteria (FJLB) preparation

The FJLB was prepared from Mulato II grass or Verano stylo before harvesting; 200 g of fresh grass was macerated with 600 ml of distilled water using a blender. The macerate was filtered and 50 ml of the filtrate was put into each flask. These filtrates in the flask were treated with glucose at the rate of 2% of volume and incubated at 30 °C for 2 days.

4.3.4 Feed and Animals

Eight male ruminally fistulated crossbred Boer × Anglo-Nubian goats (approximately 22.8 kg average body weights) were used as randomly assigned as 2 × 2 factorial arrangements in 4 × 4 replicated Latin square design to receive four dietary treatments. Dietary treatments were two species of forage (grass and legume) and two formation of roughage (fresh and silage). Each period length was 28 day of which the first 7 day used as adjustment period to the experimental diets. During each period, animals were received concentrate at 1.5% of BW and *ad libitum* roughage. Additionally, all goats were housed individually in well ventilation and shed having individual feeding and watering arrangements. All goats were provided by clean

drinking water at all time. They were dewormed at the beginning by Ivomectin injection, treated against intestinal helminthes, and intramuscular injected with vitamin AD_3E. The experimental treatments are follows as:

Treatment 1: Fresh Mulato II grass

Treatment 2: Fresh Verano stylo

Treatment 3: Mulato II grass silage

Treatment 4: Verano stylo silage

The goats were weighed every 28 days at the end of each experimental period. Individual daily DM intake was recorded

4.3.5 Metabolism trial

One metabolism trial of six days collection was conducted for nutrient utilization in goats. The metabolic cages were specially designed with a facility for separate collection of feces and urine. The animals were kept in metabolic cages for 3 days, prior to actual collection of 7 days to acclimatize the animals to the new surroundings. The appropriate aliquots of feed offered, residue left, feces were preserved animal wise for the day for chemical analysis. Body weight of the animals was recorded before and after the metabolism trials.

Measurement data of feed offer and residue were obtained. For further analysis, about 10% of feces (fresh weight) from each goat was taken daily and accumulated in a deep freezer at -20 °C until the end of the experiment. Feces from the 7 days were thoroughly mixed and then samples were taken and dried at 60 °C for 12 hours. Dried samples were ground with a mortar and pestle, the determination of dry matter (DM) was done by drying at 105 °C for 24h, ash content was assayed by incinerating samples at 550 °C, and organic matter (OM) could therefore be obtained.

Nitrogen (N) was determined by the Macro Kjeldahl technique (AOAC, 1985) and crude protein calculated as N × 6.25. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed followed the procedure described by Goering and Van Soest (1970).

4.3.6 Chemical analysis

For forage quality at harvest, a measured area of 1 m^2 was hand clipped and weighed. Each subsample was dried to determine DM content, then grounded to pass through a 1 mm mesh screen and analyzed for chemical composition. Total N was determined using the Kjeldahl method and crude protein (CP) was calculated by multiplying the N content by 6.25. Ether extract (EE) and ash contents were quantified by AOAC (1990). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) determined by the methods described by Goering and van Soest (1970).

4.3.7 Rumen fermentation and blood urea nitrogen in plasma

Rumen fluid samples from all goats were collected through ruminal fistula at 0 (prior to feeding), 2 and 4 hours post at the end of each period. It was strained through 4 layers of cheese cloth and pH measured immediately using a glass electrode pH meter. The rumen fluid was then acidified with H_2SO_4 (10%, v/v) and stored at -20 °C for subsequently quantifying NH_3-N and volatile fatty acids (VFAs) concentration. The NH_3-N were determined using distillation method according to the Kjeldahl method. The acetic acid (AA), propionic acid (PA), butyric acid (BA) and total VFAs were determined by high performance liquid chromatography (HPLC, Shim-pack SCR-102H, 300 mm × 8.0 mm i.d.; column temperature, 40 °C; flow rate, 0.8 ml/min, Shimadzu Co. Ltd., Kyoto, Japan).

Blood samples were taken from the jugular vein at 0 (prior to feeding), 2 and 4 hours post feeding. Then, the blood samples were prior to plasma separation by centrifugation (3,000 xg for 15 min) and plasma samples were then stored at -20 °C for determining blood urea nitrogen (BUN) concentration.

4.3.8 Fatty acid methyl ester of oil samples

The samples of fresh forage, forage silage and rumen fluid were immediately frozen at -20 °C until analysis. All samples were prepared for FA analysis by gas chromatography (GC) of fatty acid methyl ester (FAME). The lipid was extracted from the sample using the chloroform/methanol (2/1) method procedure of Folch et al. (1957) and Methylation of samples by the procedure described by Metcalfe (1966) was used. Fatty acid composition was measured after methylation of samples and fatty acid methyl esters (FAME) were analyzed on a Perkin Elmer Auto system gas chromatograph equipped with a flame-ionization detector (FID) using a capillary column (SPTM - 2560, 100 m × 0.25 mm ID, 0.20μm film). This analyzed adopted a split injection (split ratio 100:1). The GC analysis was temperature programmed, at 140 °C held at 5 min, and raise from 140 °C to 240 °C at a rate of 4 °C/min and then held 240 °C for 40 min. The injection port and detector temperatures were set at 260 °C. Helium was used as the carrier gas at a rate of 20 cm/sec. Identification of the FA was based upon retention times using standards of methyl esters. A mixture of the standards of the individual FAME was used to determine response factors. The areas of the peaks in the chromatogram were calculated and normalized using response factors. Proportions of individual FA were calculated.

4.4 Statistical analysis

Data were statistically analyzed according to 2 × 2 factorial arrangements in in 4 × 4 replicated Latin square design using the PROC GLM procedure (SAS, 1990) for grass and legume. Significant differences ($P<0.05$) among treatments were determined using Duncan's News Multiple Range Test according to Steel and Torrie (1980).

4.5 Results

4.5.1 Chemical and fatty acid composition of experimental diets

The chemical composition and FA pattern of the experimental treatments are demonstrated in the Table 4.1 and 4.2. For chemical composition, this was close to each other for the main nutrients (OM, CP, NDF, ADF, EE and Ash) in the experimental diets. The FA profile of the experimental diets mainly contained C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3. The grouped FAs in the experimental diets were mainly PUFA and SFA while monounsaturated FAs was the lowest content of total fat.

4.5.2 Growth performance, feed intake and nutrient digestibility

In Table 4.3, there was no effect ($P>0.05$) of both forage species, form of forage and their interactions on body weight change, DM intake of concentration and apparent nutrient (OM, CP, ADF, ADF and EE) digestibility, whereas there were different effect of forage ($P<0.05$) for DM intake of roughage and also total feed intake with the presence of forage form and interaction of forage species and form for some intake variables. When species effect was considered, the DM roughage intake as %BW ($P<0.01$), g/day ($P<0.05$) and g/$BW^{0.75}$ ($P<0.05$) were highest for the goats

fed on Mulato II grass silage among all experimental diets. Additionally, the effect of forage form ($P<0.05$) and interaction of species and from ($P<0.01$) were also presented. For total intake, there was also highest DM total intake for the goats fed on Mulato II grass silage among all experimental diets, which were similar effects to the intake of roughages as mentioned earlier. However, for intake of nutrients (OM, CP and EE), there were higher intake of OM ($P<0.05$), CP ($P<0.001$) and EE ($P<0.05$) for the goats fed on Mulato II grass silage and no effect of forage form and interaction of their interactions when compared with the goats fed on both fresh and silage of Verano stylo.

Table 4.1 Chemical composition of experimental treatments.

Item	Mulato II grass		Verano stylo	
	Fresh	Silage	Fresh	Silage
	---------- % DM ----------			
DM	23.78	29.73	33.10	32.02
OM	83.08	89.19	82.00	86.48
CP	8.55	6.22	9.76	10.17
NDF	65.50	63.06	64.42	56.86
ADF	45.02	35.64	35.54	28.57
EE	1.90	3.56	2.83	4.10
Ash	19.69	13.57	12.95	10.81

Table 4.2 Fatty aid profile (g/ 100 g total fat, % on fresh matter basis) of experimental treatments.

Item	Mulato II grass		Verano stylo	
	Fresh	**Silage**	**Fresh**	**Silage**
Fatty acid profile, % of total fat				
C12:0	0.07	0.06	0.08	0.07
C14:0	1.29	1.21	0.94	0.03
C15:0	1.14	1.41	0.22	0.00
C16:0	18.18	20.30	24.75	16.76
C16:1	0.04	0.82	1.22	1.41
C17:0	0.17	0.14	0.28	0.74
C18:0	3.24	3.55	6.44	5.89
C18:1n9	16.86	5.49	9.79	3.31
C18:2n6	14.33	16.42	8.58	16.82
C18:3n3	40.58	47.96	43.10	51.84
C20:0	0.05	0.04	0.05	0.27
C22:0	0.04	0.04	0.10	0.07
C24:0	0.28	0.39	0.29	0.14
SFA	24.46	27.14	33.15	23.97
MUFA	16.9	6.31	11.01	4.72
PUFA	54.91	64.38	51.68	68.66
PUFA/SFA	2.24	2.37	1.56	2.86

SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + C18:3n3.

4.5.3 Rumen fermentation and blood urea nitrogen in plasma

The effects of dietary treatments on rumen fermentation and blood urea nitrogen (BUN) in plasma have been shown in the Table 4.4. There were no effect of experimental treatments on rumen fermentation (P>0.05), except for the level of AA (P<0.05 for interaction effect) and PA (P<0.05 for forage form effect) at 4 hours and 2 hours, respectively, after feeding the experimental diets.

Table 4.3 Effect of dietary treatments on BW, feed intake and nutrient digestibility of the experimental goats.

Item	Mulato II grass		Verano stylo		SEM	S^{β}	F^{γ}	S × F
	Fresh	Silage	Fresh	Silage				
Initial weight, kg	22.33	23.50	23.17	22.10	0.482	ns	ns	ns
Final weight, kg	24.25	24.58	24.67	23.10	0.445	ns	ns	ns
Body weight change, kg	1.92	1.08	1.50	1.00	0.230	ns	ns	ns
Dry matter intake								
Concentrate								
g/day	362.81	339.38	362.81	360.00	6.343	ns	ns	ns
%kg BW	1.50	1.50	1.50	1.50				
g/kg $BW^{0.75}$	33.19	32.69	33.24	33.16	0.154	ns	ns	ns
Roughage								
g/day	218.84^{B}	382.57^{A}	246.34^{B}	217.37^{B}	13.237	*	*	**
%kg BW	0.90^{B}	1.65^{A}	1.03^{B}	0.89^{B}	0.048	**	*	***
g/kg $BW^{0.75}$	19.95^{B}	36.20^{A}	22.71^{B}	19.70^{B}	1.094	*	*	***

$^{A, B, C}$ Means followed by a different letter within the same row are significant different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean $^{\beta}$ S = effect of forage species (Mulato II grass and Verano stylo) $^{\gamma}$ F = effect of forage form (Fresh and Silage)

Table 4.3 (Cont.) Effect of dietary treatments on BW, feed intake and nutrient digestibility of the experimental goats.

Item	Mulato II grass		Verano stylo		SEM	S^{β}	F^{γ}	S × F
	Fresh	Silage	Fresh	Silage				
Total								
g/day	580.09^{B}	729.71^{A}	609.15^{B}	589.87^{B}	15.383	*	*	**
%kg BW	2.40^{B}	3.15^{A}	2.39^{B}	2.53^{B}	0.048	**	*	***
g/kg $BW^{0.75}$	53.08^{B}	69.09^{A}	55.95^{B}	53.16^{B}	1.113	*	*	***
Nutrient intake								
OM, g/day	603.21^{AB}	705.61^{A}	564.02^{B}	552.46^{B}	20.844	*	ns	ns
CP, g/day	89.22^{AB}	96.54^{A}	66.56^{C}	75.33^{BC}	2.573	***	ns	ns
EE, g/day	35.98^{AB}	38.82^{A}	27.68^{B}	29.10^{B}	1.452	*	ns	ns
Appearance digestibility, %								
OM	61.17	61.75	69.33	65.83	1.775	ns	ns	ns
CP	79.61	74.75	77.49	79.85	1.222	ns	ns	ns
NDF	83.06	79.68	80.39	82.57	1.270	ns	ns	ns
ADF	74.74	70.60	70.83	73.02	1.820	ns	ns	ns
EE	87.66	88.45	88.83	89.87	2.109	ns	ns	ns

$^{A, B, C}$ Means followed by a different letter within the same row are significant different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean $^{\beta}$ S = effect of forage species (Mulato II grass and Verano stylo) $^{\gamma}$ F = effect of forage form (Fresh and Silage)

4.5.4 Fatty acid composition of rumen fluid

The FA composition of rumen fluid collected from the experimental goats has been shown in the Table 4.5. The high proportions of FA in the rumen fluid of the goats were C16:0, C18:0 and C18:1n9. The significant studied effects were found in some FAs contained at low concentration (less than 5%). The main FA contents (C16:0 and C18:0), SFA, PUFA and PUFA/SFA ration were not significant differences (P>0.05) at all studied hours post feeding, however the proportion of C18:1n9 at all

studied hours post feeding (P<0.05) and MUFA at the start of feeding trial (P<0.05) in the rumen fluid of the goats fed on the fresh Mulato II grass was highest value.

Table 4.4 Effect of dietary treatments on rumen characteristics and blood urea nitrogen in plasma of the experimental goats.

Item	Hour[δ]	Mulato II grass		Verano stylo		SEM	S[β]	F[γ]	S × F
		Fresh	Silage	Fresh	Silage				
BUN, mg/dl	0	17.60[B]	20.50[AB]	23.38[A]	21.10[AB]	0.491	**	ns	ns
	2	25.00[B]	27.38[AB]	29.63[AB]	30.17[A]	0.662	*	ns	ns
	4	24.07[B]	26.81[AB]	29.88[A]	31.42[A]	0.835	**	ns	ns
pH	0	7.10	6.94	7.09	6.96	0.040	ns	ns	ns
	2	5.94	6.09	6.21	5.87	0.038	ns	ns	ns
	4	6.54	6.31	6.39	6.34	0.104	ns	ns	ns
NH_3-N, mg/dl	0	5.86	6.83	8.15	5.74	0.417	ns	ns	ns
	2	15.84	12.98	13.22	12.86	0.984	ns	ns	ns
	4	11.10	8.03	9.31	9.74	0.572	ns	ns	ns
Acetic acid, %Molar	0	71.09	74.73	75.38	74.56	1.116	ns	ns	ns
	2	69.27	66.50	71.75	69.20	2.619	ns	ns	ns
	4	67.34[B]	68.35[A]	68.85[A]	66.54[AB]	1.803	ns	ns	*
Propionic acid, %Molar	0	20.43	16.28	16.23	15.88	0.286	ns	ns	ns
	2	21.59[B]	25.48[A]	20.09[B]	22.44[AB]	1.246	ns	*	ns
	4	23.26	24.13	22.25	23.90	0.945	ns	ns	ns
Butyric acid,% Molar	0	8.48	8.99	8.39	9.56	0.191	ns	ns	ns
	2	9.13	8.02	8.16	8.36	0.407	ns	ns	ns
	4	9.40	7.52	8.90	9.55	0.516	ns	ns	ns
Total VFA, mmol/L	0	42.35	31.74	34.13	36.17	1.893	ns	ns	ns
	2	86.26	103.93	87.63	92.83	2.827	ns	ns	ns
	4	76.09	88.02	90.48	82.47	2.790	ns	ns	ns

[A, B, C] Means followed by a different letter within the same row are significant different (P<0.05), ns: not significant different (P>0.05), *P<0.05; **P<0.01; ***P<0.001, SEM: standard error of mean, BWC = Body weight change, BWG = Body weight gain, [β] S = effect of forage species (Mulato II grass and Verano stylo) [γ] F = effect of forage form (Fresh and Silage).

Table 4.5 Effect of dietary treatments on fatty acid composition of rumen fluid of the experimental goats.

Item	Hour[δ]	Mulato II grass		Verano stylo		SEM	S[β]	F[γ]	S × F
		Fresh	Silage	Fresh	Silage				
C12:0	0	3.39	4.08	4.00	4.09	0.257	ns	ns	ns
	2	6.37	7.68	8.39	7.18	0.323	ns	ns	ns
	4	6.95	6.31	7.90	6.47	0.435	ns	ns	ns
C14:0	0	3.07^{B}	4.56^{AB}	5.49^{A}	3.02^{B}	0.226	ns	ns	*
	2	3.87	4.52	4.88	4.63	0.299	ns	ns	ns
	4	6.95	6.31	7.90	6.47	0.435	ns	ns	ns
C14:1	0	4.36	4.39	5.23	4.39	0.258	ns	ns	ns
	2	3.77	3.22	4.51	3.65	0.279	ns	ns	ns
	4	2.97^{B}	3.74^{AB}	4.71^{A}	4.73^{A}	0.221	*	ns	ns
C15:0	0	1.51	1.67	1.40	1.69	0.052	ns	ns	ns
	2	0.97	0.84	0.89	0.83	0.050	ns	ns	ns
	4	0.93	0.82	0.89	0.82	0.081	ns	ns	ns
C16:0	0	28.35	28.83	28.68	29.92	0.472	ns	ns	ns
	2	24.38	26.95	26.47	26.31	0.404	ns	ns	ns
	4	26.33	26.82	26.69	26.52	0.306	ns	ns	ns
C17:0	0	1.32^{A}	1.14^{A}	0.77^{B}	0.77^{B}	0.099	*	ns	ns
	2	0.74	0.53	0.48	0.46	0.036	ns	ns	ns
	4	0.59	0.52	0.48	0.41	0.031	ns	ns	ns
C18:0	0	44.30	43.23	40.01	40.17	0.849	ns	ns	ns
	2	32.51	34.08	32.56	32.30	1.093	ns	ns	ns
	4	32.14	35.47	33.71	36.40	1.309	ns	ns	ns

[A, B, C] Means followed by a different letter within the same row are significantly different (P<0.05), ns: not significant different (P>0.05), *P<0.05, SEM: standard error of mean, [δ]Hour = Hour(s) after feeding, experimental diets, [β]S = effect of forage species (Mulato II grass and Verano stylo), [γ]F = effect of forage form (Fresh and Silage). SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + C18:3n3.

Table 4.5 (Cont.) Effect of dietary treatments on fatty acid composition of rumen fluid of the experimental goats.

Item	Hour[δ]	Mulato II grass		Verano stylo		SEM	S[β]	F[γ]	S × F
		Fresh	Silage	Fresh	Silage				
C18:1n9	0	14.96^{A}	8.36^{B}	10.84^{AB}	10.29^{AB}	0.919	ns	*	ns
	2	20.79^{A}	13.80^{B}	13.42^{B}	15.16^{AB}	0.967	ns	ns	*
	4	17.98^{A}	12.45^{B}	14.62^{AB}	13.23^{AB}	0.738	ns	*	ns
C18:2n6	0	0.90	1.08	1.08	1.08	0.093	ns	ns	ns
	2	2.36^{AB}	1.28^{B}	2.79^{AB}	3.33^{A}	0.310	*	ns	ns
	4	1.29	1.06	1.68	1.49	0.232	ns	ns	ns
C18:3n3	0	0.17^{AB}	0.04^{C}	0.10^{BC}	0.22^{A}	0.016	ns	ns	**
	2	0.17	0.21	0.12	0.15	0.020	ns	ns	ns
	4	0.11	0.10	0.12	0.10	0.018	ns	ns	ns
C18:3n6	0	0.52^{A}	0.10^{B}	0.10^{B}	0.09^{B}	0.061	ns	*	ns
	2	0.05	0.07	0.06	0.05	0.010	ns	ns	ns
	4	0.06	0.09	0.09	0.07	0.011	ns	ns	ns
C20:1	0	1.20	1.21	1.09	1.53	0.075	ns	ns	ns
	2	1.94^{AB}	1.39^{B}	1.21^{B}	2.38^{A}	0.145	ns	ns	*
	4	1.73	1.10	1.09	1.85	0.184	ns	ns	ns
C24:0	0	0.43	0.58	0.40	0.46	0.028	ns	ns	ns
	2	0.48	0.51	0.39	0.52	0.032	ns	ns	ns
	4	0.49	0.67	0.38	0.46	0.060	ns	ns	ns
SFA	0	82.80	83.19	79.27	80.17	0.735	ns	ns	ns
	2	69.35	74.75	73.62	72.16	1.420	ns	ns	ns
	4	72.57	73.56	74.30	75.07	1.223	ns	ns	ns

$^{A, B, C}$ Means followed by a different letter within the same row are significantly different (P<0.05), ns: not significant different (P>0.05), *P<0.05, SEM: standard error of mean, ^{δ}Hour = Hour(s) after feeding, experimental diets, ^{β}S = effect of forage species (Mulato II grass and Verano stylo), ^{γ}F = effect of forage form (Fresh and Silage). SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + C18:3n3.

Table 4.5 (Cont.) Effect of dietary treatments on fatty acid composition of rumen fluid of the experimental goats.

Item	Hour[δ]	Mulato II grass		Verano stylo		SEM	S[β]	F[γ]	S × F
		Fresh	Silage	Fresh	Silage				
MUFA	0	16.17[A]	9.86[B]	11.95[AB]	11.88[AB]	0.894	ns	*	ns
	2	24.53	18.42	18.41	21.21	1.084	ns	ns	ns
	4	21.04	17.32	19.56	19.86	0.739	ns	ns	ns
PUFA	0	1.64	1.15	1.33	1.41	0.141	ns	ns	ns
	2	2.60	1.64	2.99	2.88	0.276	ns	ns	ns
	4	1.16	1.48	1.50	1.71	0.262	ns	ns	ns
PUFA/SFA	0	0.02	0.01	0.02	0.02	0.002	ns	ns	ns
	2	0.04	0.02	0.05	0.04	0.005	ns	ns	ns
	4	0.02	0.02	0.02	0.03	0.004	ns	ns	ns

[A, B, C] Means followed by a different letter within the same row are significantly different (P<0.05), ns: not significant different (P>0.05), *P<0.05, SEM: standard error of mean, [δ]Hour = Hour(s) after feeding, experimental diets, [β]S = effect of forage species (Mulato II grass and Verano stylo), [γ]F = effect of forage form (Fresh and Silage). SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + C18:3n3.

4.6 Discussion

The proportions of chemical composition (DM, NDF, ADF, EE and Ash) of the fresh Mulato II grass were closed to those of the other grasses in different area in Turkey (Demirkus and Budag, 2010) and *Brachiaria* spp. (Herrero et al., 2001). The contents of DM, NDF, ADF, EE and ash in the fresh Verano stlo were closed to the reports in the Experiment I-I. However, the CP contents in the fresh Verano stylo was relatively low, when compared with the results in the Experiment I-I and report earlier (Hare et al., 2007) while the CP proportion in the fresh Mulato II grass was closed to the report in the Experiment I-I. This discrepancy would be by the fact that both Mulato II grass and Verano stylo were harvested in dry season, which the Verno stylo

was less resistance to dry season when compared with the Mulato II grass (Hare et al., 2003). When silages of Mulato II grass and Verano stylo were prepared with adding FJLB, the nutrient profile of the silages was improved by lowering %NDF and %ADF, indicating higher quality of silage (Kaiser et al., 2004). The decrease of NDF and ADF proportion would consequently result in changed proportion of CP, EE and ash. About concentration of FAs, the SFA, MUFA and PUFA contents in the fresh forage and forage silage were closed to those reported in the Experiment I-I and I-II, respectively. The PUFA was predominant proportion in both fresh and silage forms of studied forage. The C18:2n6 and C18:3n3 was the main proportion of PUFA in both fresh and silage forms of the two types of experimental forages. Generally, making silage with FJLB additive would preserve quality of studied forages.

The growth performance of the experimental goats was not differ among studied groups, although the value difference of body weight change is rather high. This would be explained by the effect of ruminal fistulation. In addition, the goats had already reached adult age, the growth rate would be low. The total intake per day of the goats was highest for the Mulato II grass silage, which influenced by the highest intake of roughage (Mulato II silage) as no difference for concentrate intake among treatments. The total intake of CP per day of the goats receiving the Mulato II grass silage was highest value while the apparent digestibility of CP of these goats had the lowest value. These might be partly explained the results of low growth rate of the goats.

The rumen fermentation of the goats receiving the experimental diets were changed at 2 and 4 h post feeding, but there were a few different measured parameters. Rumen pH of all goats was dramatically depressed at 2 h post feeding and then

gradually increased at 4 h after feeding. The concentrations of NH_3-N, AA, PA, BA and total VFA were increased at 2 h post feeding and subsequently were decreased or a bit increased at 4 h after feeding. On overview, the pattern changes of these results are in agreement with the reports earlier (Anbarasu et al., 2002; Chanjula et al., 2007). The highest concentration of AA at 4 h after feeding and PA at 2 h post feeding for the goats fed on the Mulato II grass silage might be the results of highest total feed intake. The BUN concentration in plasma were differ among the experimental groups at the start of feeding, which might be consequently resulted in differences of BUN concentration at 2 and 4 h post-feeding. However, the pattern of the BUN concentration in circulation (Anbarasu et al., 2002; Chanjula et al., 2007; Darlis et al., 2000), which some of values were in the normal range (13-26 mg/dl) of the report in Thailand (Rattana et al., 2011) whereas the rest of them were in the range reported earlier (Kohn et al., 2005). These would be explained by the variation of goats and feed.

The FA composition of rumen fluid was dramatically changed, when compared with the FAs contained in the experimental diets. The proportion of C18:2n6 and C18:3n3 were extremely reduced while the proportion of C18:0 was greatly augmented at 2 and 4 h post feeding. The proportions of C16:0 and C18:1n9 were increased when compared with those in the diet. When compared the FA composition between before and after feeding the studied diet, the FA profile of rumen fluid was largely changed at 2 h post feeding and then the FA pattern was tended to be balanced to the proportion at the start. These results would explained by the microbial hydrogenation in the rumen of the goats, leading to the conversion of C18:2n6 and C18:3n3 into C18:0 due to the high toxicity of PUFA to some rumen microorganisms

as reported by several research work (Arvidsson et al., 2009; Boufaïed et al., 2003; Buccioni et al.; Chilliard et al., 2001; Chilliard et al., 2007; Lourenço et al., 2010; Woods and Fearon, 2009). From these results in this study, it implies that microbial hydrogenation in rumen (biohydrogenation) of goats was found after receiving PUFAs. There were also illustrated that dietary PUFA contents had no effect on FA profile of rumen fluid of goats, because the microorganism in rumen of the goats did hydrogenation to balance appropriate FA proportion. Thus, suitable quantity of dietary PUFA supplementation should be given awareness and further investigated. The current study could not demonstrate the effect of dietary FAs on metabolism and deposition of FAs after feeding, but it could confirm that PUFAs were hydrogenated in the rumen of goats.

In general, feeding silage of Mulato II grass and Verano stylo to meat goats had no difference effect on feed intake, rumen fermentation and FA pattern in rumen fluid, when compared with feeding the goats with fresh form of the forages. Therefore, silage making from Mulato II grass and Verano stylo with adding FJLB would an alternative for preserving quality of the forages and could be used for roughage source for meat goats, especially during the shortage of fresh forage.

4.7 Conclusion

Mulato II grass and Verano stylo ensilage with fermented juice of epiphytic lactic acid bacteria (FJLB); as an additive had no effect on feed intake, rumen fermentation and FA pattern in rumen fluid of meat goats. Silage making from Mulato II grass and Verano stylo with adding FJLB would preserve quality of the forages for meat goats.

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CHAPTER V

EXPERIMENT III

STUDY ON EFFECTS OF MULATO II GRASS AND VERANO STYLO WITH THREE FEEDING REGIMES ON FEED INTAKE, NUTRIENT DIGESTIBILITY, RUMEN FERMENTATION, CARCASS TRAITS, MEAT QUALITY AND MUSCLE FATTY ACID PROFILE IN MEAT GOATS

5.1 Abstract

This study was aimed to study effect of Mulato II Grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*) and Verano Stylo (*Stylosanthes hamata*) with 3 feeding regimes, cut-and-carry, silage and grazing, on feed intake, nutrient utilization, rumen fermentation, carcass traits, meat quality and fatty acid composition of muscle and fat in meat goats. Plant materials were sown in a series of 10 plots (5 for each) and then were harvested for making silage prepared by adding fermented juice of epiphytic lactic acid bacteria (FJLB) and for feeding trial at 45 days after re-growth. Silage making of both forages was allowed to be fermented for 80 days at room temperature. Thirty male goats, crossbred Boer × Anglo-Nubian, were used in 2 × 3 factorial arrangements in completely randomized design. Dietary treatments were two forage sources and three feeding regimes (cut-and-carry, silage and grazing) for

each forage source. The forages were fed *ad libitum* while concentrate were offered at level of 1.5% of body weight as basal diet. The goats in the treatment of grazing regime were raised by grazing in the first plot of Mulato II grass or Verano stylo for 10 days. The forages for the goats in the experimental group of cut-and-carry feeding regime were cut from the same plot, which was used for grazing. Three goats were randomly chosen from each group and were slaughtered for determining carcass traits and meat quality. The samples of *longissimus dorsi* muscle and peritoneal fat of the meat goats were analyzed for FA composition. The results showed that BW change was influenced by feeding regime ($P<0.01$) with the presence of interaction of forage sources (S) and feeding regimes (R) ($P<0.05$). Total feed intake (g/day, g/BW0.75/day, % g/BW/day) of the goats raised by grazing Verano stylo was higher ($P<0.01$) than those in the other groups. The digestibility of OM, CP, ADF and NDF were not affected ($P>0.05$) by experimental treatments. There was found effect of forage regimes ($P<0.05$) for the digestibility of EE, which the highest fat digestibility value found in the group of the goats grazing Mulato II grass. The BUN concentration in plasma was highest ($P<0.05$) for the goats fed by Verano stylo silage at 0 and 4 hours post feeding while no difference at 2 hours post feeding. The rumen pH of the goats raised by grazing Mulato III grass and Verano stylo at the start of feeding was lower ($P<0.001$) than the other treatments. There was no effect ($P>0.05$) of the experimental treatments on the total VFA concentration at the start of feeding while the goats raised by grazing Verano stylo had highest value at 2 hours post feeding. There was no influence ($P>0.05$) of the experimental treatments among all studied groups at 2 hours after feeding while the goats fed by Mulato II grass as cut-and-carry had highest value of cellulolytic bacteria in rumen at 4 hours after feeding. There were

no effects (P>0.05) of the experimental treatments on total bacteria concentration in rumen of the studied goats at 2 and 4 hours after feeding. The concentration of protozoa in rumen of the goats at the start of feeding was not differ (P>0.05) among the experimental treatments. The meat goats raised by grazing Mulato II grass and Verano stylo had lower values of the dressing percentage while these goats had higher values of carcass length and loin eye area. The meat pH_{45min} and $pH_{ultimate}$ of the goats was rather low (P<0.01) for the goats raised by grazing Mulato II grass and Verano stylo. The meat $color_{45min}$ (L*, a* and b*) of the goats was not influenced by forage sources and feeding regimes. The experimental treatments had no effect on $color_{ultimate}$ for L* and a*, but the b* value of meat was highest for the goats fed by Mulato II grass silage. The meat of the goats raised by grazing Mulato II grass and Verano stylo had high in %drip loss. There were no effect (P>0.05) of forage sources and feeding regimes on shear force values. The EE content was lowest (P<0.05) in meat of the goats fed by Verano stylo as cut-and-carry. The main FAs of *longissimus dorsi* muscle in the studied goats are C16:0, C18:0, C18:1n9 and C18:2n6. The proportion of conjugated linoleic acid (CLA) was lowest (P<0.05) in the muscle of the goats fed by silage of Mulato II grass and Verano stylo. The ratio of PUFA/SFA was found in the muscle of the goats fed by Verano stylo as cut-and-carry. The proportion of n-6 and n-3 FAs was highest (P<0.001) in the muscle of the goats offered by Verano stylo as cut-and-carry and grazing. The ratio of n-6:n-3 was high (P<0.001) in the muscle of the goats received the silage of Mulato II grass and Verano stylo. The major FAs of peritoneal fat in the studied goats are C16:0, C18:0 and C18:1n9 while the C18:2n6 contents were remarkably depressed. The ratio of n-6:n-3 was high for the peritoneal fat of the goats received the Verano stylo as cut-and-carry and silage. In conclusion,

two forage sources with three feeding regimes, cut-and-carry, silage (prepared by adding FJLB) and grazing could be used as forage feeding without effect on concentration of BUN in plasma, rumen characteristics and microorganism population. Cut-and-carry and grazing feeding regime for both Mulato II grass and grazing Verano stylo could be applied to offer forages to meat goats with good responds of the meat goats; high growth rate, high lean yield, acceptable pH and tenderness and low fat contents with appropriate ratio of n-6:n-3 in *longissimus dorsi* muscle. However, the weaknesses of silages of both forages are low growth rate, low lean yield and high n-6:n-3 ratio in the meats and the Verano stylo offered as cut-and-carry gave low growth rate and lean yield of the meat goats.

Key Words: Forage sources, Feeding regimes, Rumen ecology, Fatty acids, Meat goats

5.2 Introduction

Over the past few years, meat from goats has gained acceptance around the world mainly because it is leaner than beef and mutton (Mahgoub et al., 2002) and has low cholesterol content (Naud'e and Hofmeyr, 1981). Moreover, goat meat is a good source of desirable fatty acids (FAs) because goats deposit higher amounts of polyunsaturated fatty acids (PUFAs) than other ruminants (Banskalieva et al., 2000; Mahgoub et al., 2002). Interest in the study of FAs, particularly the total quantity of saturated (SFA) and PUFA in muscle and adipose tissues, is mainly aimed at understanding their role in affecting human health (Banskalieva et al., 2000).

More research is needed to explore the various factors that may influence FA profiles of goat-meat products.

Hitherto, there is increasing interest in sustainable animal production systems as well as a concern for healthy, safe meat and milk products (Zervas et al., 1999). Grass- and legume based systems, can provide a good alternative to indoor ruminant production systems in order to use natural resources and to provide different meat required by consumers (Grunert et al., 2004) and, additionally, to decrease production costs (Zervas et al., 1999). Moreover, grazing ruminants are often considered of higher general quality (Ådnøy et al., 2005). In grazing animals, despite the hydrogenating effect of the rumen microorganisms, a part of essential C18:3 FA originating from the grass escapes the saturation (Wood et al., 2004) and increases the concentration of meat n-3 PUFA, compared to grain feeding (Aurousseau et al., 2004; Gatellier et al., 2004). Lambs reared under grazing conditions have greater muscle/total fat ratio (Joy et al., 2008) and higher concentration of n-3 PUFA (Wood et al., 1999). However, the production of SFA is also higher and the PUFA/SFA ratio becomes less healthy. Joy et al. (2008) compared the effects of grazing vs. concentrate-fed lambs on milk FA percentages and detected differences only the first month of lactation. Other research reported that pasture is a richer source of PUFAs than silage (Harfoot and Hazlewood, 1997). Silage making is an alternative to preserve roughage for use during constraint of forages, especially during dry season. Howevere, FAs, especially α-linolenic acid in silage may decrease, probably when undesirable fermentations occur (Lough and Anderson, 1973). In the meantime, Jonhson et al. (2010) reported the percentage of SFA, MUFA, PUFA, n-6, and n-3 FAs in *longissmus dorsi* muscle was not impacted by forage-based diet and low level concentrate. Thus, feeding regimes for forages to

meat goats are also crucial factors affecting growth rate, carcass traits and meat quality of the meat goats as concentrate taking important part of goat production (Ryan et al., 2007; Sahlu et al., 2004). Cut-and-carry method may a feeding regime giving the closed results with grazing pasture. However, there is less information for effects of feeding regimes on production, rumen fermentation and FAs profile in meat goats.

The objective of this study was to investigate the effect of forage sources and feeding regimes on feed intake, nutrient digestibility, carcass traits, meat quality and FA composition in *longissimus dorsi* muscle and peritoneal fat of meat goats.

5.3 Materials and methods

5.3.1 Plant materials

The grass and legume used for feeding experimental meat goats in the current study were Mulato II grass (*Brachiaria ruziziensis* × *B. brizantha* × *B. decumbens*) and Verano Stylo (*Stylosanthes hamata*), respectively. A series of 10 plots (each 20 × 20 m) was prepared with surrounding defense and watering system. The 10 plots were allocated into two groups equally; 5 plots for sowing Mulato II grass and the other 5 plots for sowing Verano stylo. There was no fertilizer used and the forages were sown on June 2010 at Institute of Agricultural Technology, Suranaree University of Technology, Nakhon Ratchasima.

5.3.2 Fermented juice of epiphytic lactic acid bacteria (FJLB) preparation

The FJLB was prepared from Mulato II grass or Verano stylo before harvesting; 200 g of fresh grass was macerated with 600 ml of distilled water using a blender. The macerate was filtered and 50 ml of the filtrate was put into each flask.

These filtrates in the flask were treated with glucose at the rate of 2% of volume and incubated at 30 °C for 2 days.

5.3.3 Silage making

After forage harvesting, the experimental forages were immediately chopped into 1-2 cm-length pieces. Then, fermented juice of epiphytic lactic acid bacteria (FJLB) was added at 1% of fresh matter as a silage additive, while no additive added for the control grass and legume silages. Approximate 80 kg of grass or legume were compressed in a plastic bulk. Thirty replicated plastic bulks per each treatment were prepared and allowed to be fermented for 80 days at room temperature.

5.3.4 Animals and feeding management

Thirty male goats, crossbred Thai native × Anglo-Nubian, were used in 2 × 3 factorial arrangements in CRD. Dietary treatments were two sources of forage and three feeding regimes (cut-and-carry, silage and grazing) for each forage source. The forages were fed *ad libitum*. All goats were fed concentrate at level of 1.5% of body weight as basal diet and clean drinking water was provided. Period length was last for 120 days of which the first 7 days used as adjustment period to the experimental diets. The goats were randomly allocated to 6 experimental groups of 5 each. The experimental groups were; 1) Mulato II grass with cut-and-carry feeding, 2) Mulato II grass silage, 3) Mulato II grass by grazing, 4) Verano stylo with cut-and carry feeding, 5) Verano stylo silage and 6) Verano stylo by grazing.

After Mulato II grass and Verano stylo reach 60 day-old, they were cut to allow re-growth for 45 days and then cut for making silage of Mulato II grass and Verano stylo with FJLB addition. For the feeding regimes of cut-and-carry and grazing, the forages of the 5 plots were provided for the cut-and-carry and grazing

regime as rotational grazing management. The goats in the treatment of grazing regime were raised by grazing in the first plot of Mulato II grass or Verano stylo for 10 days. After that these goats were moved to the next plot and spent 10 days per plot until the end of 120 day study. At the end of each 10 days, the forages in the plot were cut for regrowth throughout the plot. In the meantime, the forages for the goats in the experimental group of cut-and-carry feeding regime were cut from the same plot, which was using for grazing.

5.3.5 Metabolism trial

One metabolism trial of six days collection was conducted for nutrient utilization in goats. The metabolic cages were specially designed with a facility for separate collection of feces and urine. The animals were kept in metabolic cages for 3 days, prior to actual collection of 7 days to acclimatize the animals to the new surroundings. The appropriate aliquots of feed offered, residue left, feces were preserved animal wise for the day for chemical analysis. Body weight of the animals was recorded before and after the metabolism trials.

Measurement data of feed offer and residue were obtained. For further analysis, about 10% of feces (fresh weight) from each goat was taken daily and accumulated in a deep freezer at -20 °C until the end of the experiment. Feces from the 7 days were thoroughly mixed and then samples were taken and dried at 60 °C for 12 hours. Dried samples were ground with a mortar and pestle, the determination of dry matter (DM) was done by drying at 105 °C for 24h, ash content was assayed by incinerating samples at 550 °C, and organic matter (OM) could therefore be obtained. Nitrogen (N) was determined by the Macro Kjeldahl technique (AOAC, 1985) and crude protein calculated as N*6.25. Neutral detergent fiber (NDF) and acid detergent

fiber (ADF) were analyzed followed the procedure described by Goering and Van Soest (1970).

5.3.6 Rumen fermentation and blood urea nitrogen in plasma

After 120 days of the experiment, the rumen contents was collected before feeding (0 hour), 2 and 4 hours after feeding using a stomach tube attached to a suction pump, pH measured immediately using a glass electrode pH meter. After recording pH, aliquot of the samples were strained through 4 layers of cheese cloth. The rumen fluid was then acidified with H_2SO_4 (10%, v/v) and stored at -20 °C for subsequently quantifying NH_3-N and volatile fatty acids (VFAs) concentration. The NH_3-N were determined using distillation method according to the Kjeldahl method. The acetic acid (AA), propionic acid (PA), butyric acid (BA) and total VFAs were determined by high performance liquid chromatography (HPLC, Shim-pack SCR-102H, 300 × 8.0 mm i.d.; column temperature, 40 °C; flow rate, 0.8 ml/min, Shimadzu Co. Ltd., Kyoto, Japan).

Blood samples were taken from the jugular vein at 0 (prior to feeding), 2 and 4 hours post feeding. Then, the blood samples were prior to plasma separation by centrifugation (3,000 xg for 15 min) and plasma samples were then stored at -20 °C for determining blood urea nitrogen (BUN) concentration.

5.3.7 Slaughter procedure and carcass characteristics

At the end of 120 day fattening period, 3 goats were randomly chosen from each group and were stunned with a captive-bolt pistol at the experimental slaughter unit at Suranaree University of Technology (SUT). After slaughtering, non-carcass components (head, skin, feet, trachea and lungs, liver, heart, spleen, pancreas, gastro-intestinal tract, diaphragm and testicles) were removed from carcass and weighed, and

then the rest of component was hot carcass weighed. Hot carcass included kidneys and perinephric-pelvic fat as described by Colomer-Rocher et al. (1987). Cold carcass weight was obtained after chilling the hot carcass at 4 °C for 24 hours. Dressing percentage was calculated as hot carcass weight divided by slaughter BW. Carcass length was measured from the top point of shoulder (anterior part of scapula) to the tuber ischium (pelvic bone) and loin eye area were measured from both sides of each carcass, at the 12^{th} and 13^{th} rib using a one-centimeter grid (each dot on the grid represents 0.1 square inches of measurement). Lean weight was estimated from formula: Lean weight (kg) = -1.09 + (0.8 × Cold carcass weight (kg)); R^2 = 0.98 (Hopkins-Shoemaker, 2006). Lean percentage was calculated as lean weight (kg) × 100 / hot carcass weight (kg).

5.3.8 Meat quality analysis

Instrumental meat quality characteristics investigated in the current study were carcass pH, drip loss (%), water holding capacity (%), shear force (kg) and meat color (L*, a*, b*). Carcass pH was measured at 45 min after slaughter (pH_{45}) and at 24 hour post-slaughter (pH_{24}) using a digital pH meter. The pH measurement was performed directly on *longissimus thoracic* muscle between 12^{th} and 13^{th} thoracic vertebrae.

The *longissimus dorsi* muscle was removed from the right side of the carcass at 24 h post-mortem in order to assess instrumental meat quality characteristics. *Longissimus thoracic* muscle between 6^{th} and 13^{th} ribs was used for shear force determination, while samples from the *longissimus lumborum* muscle used for meat color and drip loss measurements.

Meat color was measured after 1 h storage (first measurement) and finally after 24 h storage (second measurement) on cut surface of 2.5 cm thick samples from fat-free area. During the storage period, samples were kept at 4 °C in a polystyrene tray and over wrapped with oxygen permeable PVC film to allow blooming. Nine color measurements performed from each sample and color coordinate value determined by calculating the average of these nine measurements. Color was evaluated using the CIELAB color space. L* (lightness), a* (redness) and b* (yellowness) using a Minolta CM-2006 d spectropho- tometer (Konica Minolta Holdings, Inc, Osaka, Japan).

Drip loss was determined using the method described by Honikel (1998). Briefly, meat samples were weighed and then suspended in an inflated polyethylene bag without any contact with the bag. After a 24 hours storage period at 4 °C, the samples were gently dried with paper towels, and reweighed. Drip loss (%) was estimated by the ratio of weight loss to initial sample weight.

Shear Force was measurement after 24 hours at 4 °C. Six samples (2.5 cm diameter, 2.0 cm length) per treatment were sheared perpendicular to fibre direction using an Instron 4501 Universal Testing Machine (Instron Corp., Canton, USA) equipped with a Warner Bratzler shear force cell. Load cell and cross-head speed were 5 KN and 1.0 cm length, 100 mm min^{-1}, respectively. The maximum peak recorded was taken as the shear force.

5.3.9 Fatty acid methyl ester of oil samples

The samples of *longissimus dorsi* muscle and peritoneal fat of the meat goats were collected and were immediately frozen at -20 °C until analysis. All samples were prepared for FA analysis by gas chromatography (GC) of fatty acid methyl ester

(FAME). The lipids were extracted from the forages using the chloroform/methanol (2/1) method procedure of Folch et al. (1957). For quantification of CLA isomers, lipids extracted from samples were methylated (sodium methoxide) following the method of Li and Watkins (1998). Methylation of samples by the procedure described by Metcalfe (1966) was used. Fatty acid composition was measured after methylation of samples. Fatty acid methyl esters were analyzed on a Perkin Elmer Auto system gas chromatograph equipped with a flame-ionization detector (FID) using a capillary column (SPTM - 2560, 100 m x 0.25 mm ID, 0.20 μm film). This analyzed adopted a split injection (split ratio 100:1). The GC analysis was temperature programmed, at 140 °C held at 5 min, and raised from 140 °C to 240 °C at a rate of 4 °C/min and then held 240 °C for 40 min. The injection port and detector temperatures were set at 260 °C. Helium was used as the carrier gases at a rate of 20 cm/sec. Identification of the FA were based upon retention times using standards of methyl esters. A mixture of the standards of the individual FAME was used to determine response factors. The areas of the peaks in the chromatogram were calculated and normalized using response factors. Proportions of individual FA were calculated.

5.4 Statistical analysis

Data were statistically analyzed according to 2 × 3 factorial in CRD using the PROC GLM procedure (SAS, 1990). Significant differences ($P<0.05$) among treatments were determined using Duncan's News Multiple Range test according to Steel and Torrie (1980).

5.5 Results

5.5.1 Chemical composition of concentrate and roughages

The composition of dietary concentrate, Mulato II grass and Verano stylo with different feeding regimes has been shown in the Table 5.1. The chemical composition of all experimental treatments was closed to each other, except for %ash. The FA profile of the experimental diets mainly contained C16:0, C18:0, C18:1n9, C18:2n6 and C18:3n3. The grouped FAs in the experimental diets were mainly polyunsaturated fatty acids (PUFA) and saturated fatty acids (SFA) while monounsaturated fatty acids (MUFA) was the lowest content of total fat.

5.5.2 Growth rate, feed intake and nutrient digestibility

The intake of concentrate and roughages has been shown in Table 5.2. There were no different of initial BW of goats ($P>0.05$). The BW change was influenced by feeding regime ($P<0.01$) with the presence of interaction of forage sources (S) and feeding regimes (R) ($P<0.05$). The intake (g/day, g/$BW^{0.75}$/day, % g/BW/day) of concentrate and roughage was high for the goats raised with grazing Verano stylo. The total intake (g/day, g/$BW^{0.75}$/day, % g/BW/day) of the goats raised by grazing Verano stylo was higher ($P<0.01$) than those in the other groups. These would be the results of high intake of concentrate and roughage. The digestibilities of OM, CP, ADF and NDF were not affected ($P>0.05$) by experimental treatments. There was found effect of forage regimes ($P<0.05$) for the digestibility of EE, which the highest fat digestibility value found in the group of the goats grazing Mulato II grass while the lowest values of fat digestibility found in the groups of the goats fed the silage of Mulato II grass and Verano stylo.

5.5.3 Rumen fermentation and blood urea nitrogen in plasma

The effects of dietary treatments on rumen characteristics, blood urea nitrogen (BUN) in plasma and rumen microorganisms have been shown in the Table 5.3. The BUN concentration in plasma was highest ($P<0.05$) for the goats fed by Verano stylo silage at 0 and 4 hours post feeding while no difference at 2 hours post feeding. The rumen pH of the goats raised with grazing Mulato III grass and Verano.

Table 5.1 Chemical composition (% on dry matter basis) and fatty aid profile (g/100g total fat, on fresh matter basis) of the experimental treatments.

Item	Mulato II grass			Verano stylo		
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing
DM, %	24.29	22.64	24.03	37.11	27.67	33.42
OM, %	90.27	87.23	88.24	77.43	85.78	91.24
CP, %	8.85	9.61	8.71	8.17	8.27	7.77
EE, %	3.83	4.23	3.84	2.10	3.00	2.59
NDF, %	65.99	62.10	67.72	62.75	55.84	64.94
ADF, %	34.67	34.43	32.69	36.88	33.78	42.64
Ash, %	9.74	12.77	11.77	22.57	14.23	8.77
Fatty acid profiles, g/100 g total fat						
C12:0	0.61	0.05	0.60	0.60	0.07	0.36
C14:0	1.25	1.21	1.66	1.71	0.94	2.23
C15:0	1.14	1.41	1.10	0.40	0.10	0.20
C16:0	18.18	19.80	17.65	24.75	16.76	23.42

SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

Table 5.1 (Cont.) Chemical composition (% on dry matter basis) and fatty aid profile (g/100g total fat, on fresh matter basis) of the experimental treatments.

Item	Mulato II grass			Verano stylo		
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing
C16:1	0.74	0.76	0.87	1.19	1.29	1.21
C17:0	0.28	0.32	0.45	0.29	0.66	0.19
C18:0	2.74	3.55	2.03	6.44	5.89	6.00
C18:1n9	16.25	5.17	15.55	9.79	3.08	9.51
C18:2n6	14.92	15.39	15.10	8.58	16.82	8.20
C18:3n3	40.58	47.96	41.43	43.11	51.50	43.93
SFA	24.58	26.79	23.90	34.56	24.86	32.69
MUFA	16.99	5.93	16.42	10.97	4.37	10.72
PUFA	55.50	63.35	56.53	51.68	68.32	52.13
PUFA/SFA	2.27	2.37	2.38	1.50	2.76	1.60

SFA: saturated fatty acid = C12:0+C14:0+C15:0+C16:0+C17:0+C18:0, MUFA: monounsaturated fatty acid = C16:1+C18:1n9 and PUFA: poly unsaturated fatty acid = C18:2n6+C18:3n3.

stylo at the start of feeding was lower ($P<0.001$) than the other treatments. There were highest value of rumen pH at 2 hours after feeding for the goats raised by Mulato II silage and grazing Verano stylo, but there was lowest value of rumen pH and highest rumen pH for the goats raised by grazing Verano stylo and Mulato II grass, respectively. At the start of the feeding, the NH_3-N concentration of the goats fed by cut-and-carry and grazing of Mulato II grass was higher ($P<0.01$) while the goats fed by grazing of Mulato II grass and Verano stylo was lower ($P<0.001$). There were no effect of the experimental treatments on NH_3-N concentration at 2 and 4 hours post feeding.

The concentration of acetic acid concentration was highest value for Verano stylo grazing and silage at 2 and 4 hours post feeding, respectively. The goats fed by Mulato II grass as cut-and-carry had the highest value of PA concentration while those fed by Verano stylo was lowest at the start of feeding. The concentration of PA for the goats raised by grazing Verano stylo had highest value at the start of feeding while the highest values of PA were found for the goats fed by Verano stylo as cut-and-carry and silage. The highest concentration of BA in rumen was found for the goats fed by Mulato II grass silage at the start of feeding, grazing Verano stylo at 2 hours post feeding and Mulato II grass & Verano stylo silage at 4 hours post feeding. When total VFA concentration was calculated, there was no effect (P>0.05) of the experimental treatments on the total VFA concentration at the start of feeding while the goats raised by grazing Verano stylo had highest value at 2 hours post feeding. The concentration of total VFA for the goats fed by Mulato II grass silage, Verano stylo as cut-and carry, and Verano silage were higher (P<0.05) than the goats fed by Mulato II grass as cut-and-carry, grazing Mulato II grass and Verano stylo.

Table 5.2 Feed intake and nutrient digestibility of the experimental meat goats fed on different forages and regimes.

Item	Mulato II grass			Verano stylo			SEM	S^{β}	F^{γ}	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
Initial BW, kg	11.50	10.75	11.68	11.20	10.60	11.38	0.310	ns	ns	ns
Final BW, kg	19.25^{B}	15.30^{C}	21.75^{A}	15.10^{C}	15.60^{C}	20.83^{AB}	0.236	*	***	*
BWC, kg	9.25^{A}	4.13^{B}	9.40^{A}	3.93^{B}	4.77^{B}	9.00^{A}	0.403	ns	**	*

$^{A, B, C, D}$ Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001; ns: not significant different (P>0.05) and SEM: standard error of mean, $^{\delta}$ Hour = Hour(s) after feeding experimental diets, $^{\beta}$ S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$ F = effect of forage form (Fresh and Silage).

Table 5.2 (Cont.) Feed intake and nutrient digestibility of the experimental meat goats fed on different forages and regimes.

Item	Mulato II grass			Verano stylo			SEM	S[β]	F[γ]	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
Dry matter intake										
Concentrate										
g/day	181.22^{B}	185.56^{B}	234.31^{A}	179.65^{B}	175.05^{B}	229.59^{A}	4.243	ns	***	ns
% g/BW/day	1.40^{AB}	1.41^{AB}	1.44^{AB}	1.34^{B}	1.31^{B}	1.47^{A}	0.015	ns	*	ns
g/BW$^{0.75}$/day	25.65^{ABC}	25.08^{BC}	29.64^{A}	23.66^{C}	26.78^{ABC}	28.90^{AB}	0.522	ns	**	ns
Roughage										
g/day	367.00^{B}	260.51^{C}	340.44^{B}	322.27^{B}	326.94^{B}	467.64^{A}	6.476	**	***	***
% g/BW/day	2.40^{AB}	1.98^{B}	2.11^{B}	2.41^{AB}	2.45^{AB}	2.87^{A}	0.060	**	ns	ns
g/BW$^{0.75}$/day	47.46^{B}	37.73^{C}	42.30^{BC}	46.06^{BC}	46.87^{B}	57.71^{A}	1.080	***	ns	*
Total										
g/day	548.80^{BC}	446.07^{D}	574.74^{B}	501.92^{C}	501.98^{C}	697.23^{A}	6.805	*	***	***
% g/BW/day	3.60^{B}	3.39^{B}	3.55^{B}	3.75^{B}	3.76^{B}	4.35^{A}	0.071	*	ns	ns
g/BW$^{0.75}$/day	71.12^{B}	64.52^{B}	71.20^{B}	71.71^{B}	71.95^{B}	87.35^{A}	1.197	*	**	ns
Apparent digestibility, %										
OM	79.13	78.72	78.25	79.97	77.73	78.66	0.882	ns	ns	ns
CP	72.62	75.22	72.90	77.28	72.62	72.66	1.323	ns	ns	ns
ADF	78.36	75.57	72.96	75.68	75.93	79.71	0.869	ns	ns	ns
NDF	83.30	81.77	79.23	79.55	77.29	79.20	0.788	ns	ns	ns
EE	72.14^{AB}	70.42^{B}	79.39^{A}	78.06^{AB}	70.29^{AB}	72.07^{AB}	1.049	ns	*	ns

$^{A, B, C, D}$ Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001; ns: not significant different (P>0.05) and SEM: standard error of mean, $^{\delta}$ Hour = Hour(s) after feeding experimental diets, $^{\beta}$ S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$ F = effect of forage form (Fresh and Silage).

5.5.4 Carcass characteristics and meat quality of the meat goats

The carcass characteristics of the meat goats have been shown in the Table 5.4. There were no differences (P>0.05) among the experimental treatment for heart

and liver, as a percent of slaughter weight (SW). The head and skin (%SW) of the meat goats fed by Mulato II grass and grazing Verano stylo were lowest ($P<0.05$). The feet (%SW) of the meat goats fed by Mulato II grass as cut-and-carry & grazing and Verano stylo grazing had lower ($P<0.05$) than those fed by the other feeding regimes. The kidney and pelvic fat ($P<0.001$) and spleen (%SW) ($P<0.05$) of the meat goats raised by grazing Verano stylo was highest and the meat goats raised with grazing Mulato II grass the second high values. The meat goats fed by Mulato II as grass as cut-and-carry & grazing and Verano stylo grazing were higher hot and cold carcass weight, and lean percentage than the other feeding regimes. The meat goats raised by grazing Mulato II grass and Verano stylo had lower values of the dressing percentage while these goats had higher values of carcass length and loin eye area.

The meat quality of the meat goats are illustrated in the Table 5.5. The meat pH_{45min} and $pH_{ultimate}$ of the goats was rather low ($P<0.01$) for the goats raised by grazing Mulato II grass and Verano stylo. The meat $color_{45\ min}$ (L*, a* and b*) of the goats was not influenced by forage sources and feeding regimes. The experimental treatments had no effect on $color_{ultimate}$ for L* and a*, but the b* value of meat was highest for the goats fed by Mulato II grass silage. The meat of the goats raised by grazing Mulato II grass and Verano stylo had high in %drip loss. There were no effect ($P>0.05$) of forage sources and feeding regimes on shear force values.

Table 5.3 Rumen characteristics and population of rumen bacteria of the experimental meat goats fed on different forages and regimes.

Hour[δ]	Mulato II grass			Verano stylo			SEM	S[β]	F[γ]	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
BUN, mg/dl										
0	13.17^{B}	17.38^{AB}	17.38^{AB}	14.20^{B}	18.90^{A}	14.25^{B}	0.536	ns	*	ns
2	19.67	23.50	20.50	20.60	25.20	23.40	0.676	ns	ns	ns
4	19.67^{B}	22.00^{B}	22.50^{B}	22.80^{B}	28.00^{A}	21.25^{B}	0.670	ns	*	ns
Rumen pH										
0	6.81^{A}	6.80^{A}	6.50^{B}	6.95^{A}	6.92^{A}	6.48^{B}	0.034	ns	***	ns
2	5.90^{A}	5.92^{A}	5.18^{B}	5.13^{B}	5.14^{B}	5.78^{A}	0.063	*	ns	***
4	6.52^{AB}	6.50^{AB}	6.68^{A}	6.26^{AB}	6.10^{B}	6.24^{AB}	0.058	**	ns	ns
NH_3 -N, mg/dl										
0	19.11^{A}	18.13^{A}	15.67^{AB}	13.49^{B}	14.38^{B}	13.73^{B}	0.485	**	ns	ns
2	20.97	16.69	21.89	17.96	18.51	18.20	1.078	ns	ns	ns
4	4.20	3.64	5.04	3.58	3.47	4.20	0.263	ns	ns	ns
Acetic acid, %Molar										
0	71.92	71.78	74.67	75.30	86.87	76.48	0.886	ns	ns	ns
2	72.01^{AB}	65.90^{B}	70.74^{AB}	71.52^{AB}	65.37^{B}	66.48^{A}	1.418	ns	*	ns
4	80.85^{BC}	75.71^{AB}	79.39^{C}	74.16^{AB}	76.16^{A}	78.35^{C}	1.480	*	***	ns
Propionic acid, %Molar										
0	17.69^{A}	15.19^{AB}	15.78^{AB}	15.57^{AB}	7.46^{C}	15.07^{BC}	0.331	*	*	ns
2	18.47^{B}	24.59^{B}	21.86^{AB}	21.13^{AB}	26.84^{AB}	23.94^{A}	0.681	*	ns	ns
4	14.39^{BC}	15.21^{AB}	14.68^{C}	18.55^{A}	15.59^{A}	15.3^{C}	0.562	*	***	ns
Butyric acid, %Molar										
0	10.39^{AB}	13.03^{A}	9.55^{AB}	9.13^{AB}	5.67^{C}	8.45^{BC}	0.302	** *	ns	*
2	9.53^{AB}	9.51^{B}	7.4^{B}	7.35^{B}	7.79^{B}	9.57^{A}	0.321	ns	ns	*
4	4.76^{B}	9.08^{A}	5.93^{B}	7.28^{A}	8.25^{A}	6.36^{B}	0.280	*	***	ns
Total VFA, mmol/L										
0	34.61	34.87	30.17	34.78	23.95	24.17	1.341	ns	ns	ns
2	50.20^{B}	45.78^{B}	53.30^{AB}	55.75^{AB}	49.12^{B}	68.06^{A}	2.055	ns	*	ns
4	37.10^{B}	54.39^{A}	23.06^{B}	54.23^{A}	61.41^{A}	31.98^{B}	2.160	*	***	ns

$^{A, B, C, D}$ Means followed by a different letter within the same row are significant difference: *P<0.05; **P<0.01; ***P<0.001; ns: not significant difference (P>0.05) and SEM: standard error of mean, ^{δ} Hour = Hour(s) after feeding experimental diets, ^{β} S = effect of forage sources (Mulato II grass and Verano stylo), ^{γ} F = effect of forage form (Fresh and Silage)

5.5.5 Chemical and fatty acid composition of *longissimus dorsi* muscle and fatty acid profile of peritoneal fat

The chemical and fatty acid composition of *longissimus dorsi* muscle of the meat goats has been demonstrated in the Table 5.6. The contents of moisture and ash were not affected (P>0.05) by the studies forage sources and feeding regime. The *longissimus dorsi* muscle of the goats fed by Mulato II grass as cut-and-carry contained highest CP content (P<0.01) while the EE content was lowest (P<0.05) in meat of the goats fed by Verano stylo as cut-and-carry. In general, the main FAs of *longissimus dorsi* muscle in the studied goats are C16:0, C18:0, C18:1n9 and C18:2n6. There were small increases the proportion of C14:0, C17:0, C17:1, C20:3n6, C20:4n6, C22:2 and C22:6n3 whereas the proportions of C15:0 and C16:1 decreased, when compared with the FA content in the experimental diets. There were no influences of forage sources and feeding regimes on concentration of C16:0 and C18:3n3 in the muscle. The proportion of conjugated linoleic acid (CLA) was lowest (P<0.05) in the muscle of the goats fed by silage of Mulato II grass and Verano stylo. When grouped FAs was calculated, the MUFAs proportion was not affected (P>0.05) by the experimental treatments while the concentration of SFA was high for the muscle of the goats fed by Mulato II grass as cut-an-carry and Verano stylo silage and the highest value of PUFAs proportion and the ratio of PUFA/SFA was found in the muscle of the goats fed by Verano stylo as cut-and-carry. The proportion of n-6 and n-3 FAs was highest (P<0.001) in the muscle of the goats offered by Verano stylo as cut-and-carry and grazing. The ratio of n-6:n-3 was high (P<0.001) in the muscle of the goats received the silage of Mulato II grass and Verano stylo.

The FA composition of peritoneal fat of the meat goats has been shown in the Table 5.7. The major FAs of peritoneal fat in the studied goats are C16:0, C18:0 and C18:1n9 while the C18:2n6 contents were remarkably depressed. There was small increase the proportion of C14:0, C16:1 and C17:0, when compared with the FA content in the experimental diets. There was no influence of forage sources and feeding regimes on concentration of C16:0, C18:0, C18:1n9 and C18:3n3 in the peritoneal fat. The highest value of the C18:2n6 proportion in peritoneal fat of the goats raised by grazing Verano stylo. The proportion of CLA in peritoneal fat had highest value for the goats raised by grazing Mulato II grass. When grouped FAs was calculated, the SFAs and MUFAs was not affected ($P>0.05$) by the experimental treatments while the concentration of PUFA was high for the peritoneal fat of the goats raised by grazing Mulato II grass and Verano stylo. The proportion of n-6 FAs was highest value for the peritoneal fat of the goats raised by grazing Verano stylo while the proportion of n-3 FAs were highest in the peritoneal fat of the goats offered by Mulato II grass as grazing. The ratio of n-6:n-3 was high for the peritoneal fat of the goats received the Verano stylo as cut-and-carry and silage.

Table 5.4 Carcass characteristics of the meat goats fed on different forage and regimes.

	Mulato II grass			Verano stylo			SEM	S^{β}	F^{γ}	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
Slaughter weight, kg	18.25^{B}	14.28^{C}	20.75^{A}	14.10^{C}	14.60^{C}	19.83^{AB}	0.235	*	***	**
Body component, % of slaughter weight										
Head	8.55^{A}	9.46^{A}	5.35^{B}	9.20^{A}	9.42^{A}	4.55^{B}	0.113	ns	***	*
Skin	12.16^{A}	10.64^{AB}	7.10^{C}	10.22^{B}	11.28^{AB}	6.61^{C}	0.203	ns	***	ns
Heart	0.54	0.75	0.55	0.66	0.67	0.66	0.025	ns	ns	ns
Liver	1.67	2.49	1.78	1.55	2.11	1.88	0.110	ns	ns	ns
Lungs	0.92^{B}	1.09^{A}	0.90^{B}	0.98^{AB}	1.08^{A}	0.89^{B}	0.076	ns	*	*
Feet	2.63^{AB}	3.70^{A}	1.49^{C}	3.31^{AB}	3.01^{AB}	2.45^{BC}	0.133	ns	***	ns
Kidney and pelvic fat	0.27^{C}	0.39^{B}	0.46^{AB}	0.33^{BC}	0.33^{BC}	0.52^{A}	0.014	ns	***	ns
Spleen	0.20^{B}	0.24^{AB}	0.24^{AB}	0.17^{B}	0.19^{B}	0.27^{A}	0.008	ns	*	ns
Carcass characteristics										
Hot carcass weight, kg	8.23^{A}	6.43^{B}	7.78^{A}	6.00^{B}	5.96^{B}	7.78^{A}	0.132	*	**	*
Cold carcass weight, kg	7.76^{A}	6.00^{B}	7.20^{A}	5.54^{B}	5.49^{B}	7.26^{A}	0.134	*	**	*
Dressing percentage, %	45.14^{A}	44.92^{A}	37.48^{B}	40.79^{AB}	40.43^{AB}	39.24^{B}	0.662	ns	*	ns
Carcass length, cm	64.00^{C}	57.67^{D}	122.00^{A}	56.85^{D}	53.67^{D}	115.67^{B}	0.081	ns	***	ns
Loin eye area, cm^2	6.81^{ABC}	5.54^{BC}	7.56^{AB}	4.26^{C}	4.67^{C}	8.39^{A}	0.328	ns	**	ns
Lean, %	63.02^{A}	58.06^{B}	61.73^{A}	56.63^{B}	56.54^{B}	61.89^{A}	0.456	ns	**	ns

$^{A, B, C, D}$ Means followed by a different letter within the same row are significant difference: *P<0.05; **P<0.01; ***P<0.001; ns: not significantly different (P>0.05) and SEM: standard error of mean, $^{\beta}$S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$F = effect of forage form (Fresh and Silage).

Table 5.5 Meat quality characteristics for *longissimus dorsi* muscle of the meat goats fed on different forages and regimes.

Item	Mulato II grass			Verano stylo			SEM	S^{β}	F^{γ}	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
$pH_{45\ min}$	6.94^{A}	7.20^{A}	6.61^{AB}	7.10^{A}	7.17^{A}	5.99^{B}	0.085	ns	**	ns
$pH_{ultimate}$	5.91^{AB}	5.89^{AB}	5.41^{B}	5.99^{AB}	6.11^{A}	5.40^{B}	0.073	ns	**	**
$Color_{45\ min}$										
L* (lightness)	44.61	46.83	48.27	49.75	47.77	47.73	0.604	ns	ns	ns
a* (redness)	11.41	11.67	12.05	12.70	12.24	11.03	0.466	ns	ns	ns
b* (yellowness)	0.70	0.33	0.03	0.77	0.13	1.05	0.335	ns	ns	ns
$Color_{ultimate}$										
L* (lightness)	46.08	47.16	50.29	48.74	50.44	50.63	0.558	ns	ns	ns
a* (redness)	14.51	14.47	12.89	14.71	14.88	12.49	0.409	ns	ns	ns
b* (yellowness)	3.88^{B}	5.90^{A}	4.38^{AB}	3.26^{B}	4.91^{AB}	5.14^{AB}	0.229	ns	*	ns
Drip loss, %	1.39^{C}	1.07^{C}	2.88^{A}	1.58^{BC}	0.98^{C}	2.06^{B}	0.081	ns	***	ns
Shear force, kg/cm^2	2.42	3.34	2.65	3.91	3.93	4.97	0.426	ns	ns	ns

$^{A, B, C}$ Means followed by a different letter within the same row are significantly different, *P<0.05; **P<0.01; ***P<0.001; ns: not significantly different (P>0.05) and SEM: standard error of mean, $^{\beta}$S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$F = effect of forage form (Fresh and Silage).

Table 5.6 Chemical composition (% on fresh matter) and fatty acid profiles (g/100 g total fat) of *Longissimus dorsi* muscle of the meat goats fed on different forages and regimes.

Item	Mulato II grass			Verano stylo			SEM	S[β]	F[γ]	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
Chemical compositions										
Moisture, %	75.16	72.00	74.43	74.38	73.27	73.36	0.404	ns	ns	ns
Ash, %	1.13	1.09	1.14	1.04	1.01	1.09	0.145	ns	ns	ns
CP, %	22.28^{A}	20.98^{AB}	20.28^{B}	19.57^{B}	19.32^{B}	19.19^{B}	0.244	**	ns	ns
EE, %	1.72^{A}	1.56^{A}	1.98^{A}	0.89^{B}	1.91^{A}	2.04^{A}	0.077	ns	*	*
Fatty acid profiles										
C12:0	0.37	0.51	0.14	0.26	0.40	0.26	0.053	ns	ns	ns
C14:0	2.87^{B}	3.28^{AB}	1.74^{C}	1.86^{C}	3.99^{A}	3.06^{B}	0.105	ns	***	***
C15:0	0.64	0.63	0.51	0.57	0.69	0.54	0.044	ns	ns	ns
C16:0	20.79	23.60	20.30	19.29	22.86	19.84	0.670	ns	ns	ns
C16:1	0.37^{B}	1.41^{A}	0.26^{B}	0.53^{B}	1.50^{A}	0.69^{B}	0.053	ns	***	ns
C17:0	2.06	2.00	1.94	1.48	1.97	1.68	0.112	ns	ns	ns
C17:1	1.92	1.34	1.77	2.31	2.04	1.56	0.171	ns	ns	ns
C18:0	24.84^{A}	18.63^{BC}	22.41^{AB}	16.27^{C}	21.56^{AB}	20.11	0.572	*	ns	ns
C18:1n9	28.18^{B}	36.64^{A}	38.33^{A}	34.26^{AB}	32.72^{AB}	34.19	0.871	ns	ns	*
C18:2n6	6.93^{A}	6.84^{A}	4.46^{B}	6.38^{AB}	7.30^{A}	6.39	0.236	ns	*	ns
CLA	0.46^{A}	0.28^{B}	0.53^{A}	0.46^{A}	0.29^{B}	0.59	0.035	ns	*	ns
C18:3n3	0.14	0.32	0.13	0.11	0.17	0.16	0.041	ns	ns	ns
C17:0	2.06	2.00	1.94	1.48	1.97	1.68	0.112	ns	ns	ns
C17:1	1.92	1.34	1.77	2.31	2.04	1.56	0.171	ns	ns	ns

$^{A, B, C, D}$ Means followed by a different letter within the same row are significant difference: *P<0.05; **P<0.01; ***P<0.001; ns: not significant difference (P>0.05) and SEM: standard error of mean, $^{\beta}$ S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$ F = effect of forage form (Fresh and Silage), SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C17:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + CLA + C18:3n3 + C20:3n6 + C20:4n6 + C22:2 + C22:6n3, CLA = Conjugated linoleic acid = *cis-9, trans-11*CLA + *trans-10, cis-12*CLA.

Table 5.6 (Cont.) Chemical composition (% on fresh matter) and fatty acid profiles (g/100 g total fat) of *Longissimus dorsi* muscle of the meat goats fed on different forages and regimes.

Item	Mulato II grass			Verano stylo			SEM	S[β]	F[γ]	S × R
	Cut-and-carry	Silage	Grazing	Cut-and-carry	Silage	Grazing				
C18:0	24.84^{A}	18.63^{BC}	22.41^{AB}	16.27^{C}	21.56^{AB}	20.11^{ABC}	0.572	*	ns	ns
C18:1n9	28.18^{B}	36.64^{A}	38.33^{A}	34.26^{AB}	32.72^{AB}	34.19^{AB}	0.871	ns	ns	*
C18:2n6	6.93^{A}	6.84^{A}	4.46^{B}	6.38^{AB}	7.30^{A}	6.39^{AB}	0.236	ns	*	ns
CLA	0.46^{A}	0.28^{B}	0.53^{A}	0.46^{A}	0.29^{B}	0.59^{A}	0.035	ns	*	ns
C18:3n3	0.14	0.32	0.13	0.11	0.17	0.16	0.041	ns	ns	ns
C20:3n6	1.36^{A}	0.23^{B}	1.31^{A}	1.25^{A}	0.33^{B}	1.56^{A}	0.057	ns	***	ns
C20:4n6	1.64^{CD}	1.21^{D}	2.71^{BC}	4.48^{A}	2.55^{BC}	3.69^{AB}	0.159	***	*	ns
C22:2	0.49	0.03	0.47	0.39	0.38	0.24	0.053	ns	ns	ns
C22:6n3	1.58^{BC}	0.40^{D}	1.30^{C}	2.67^{A}	0.44^{D}	2.40^{AB}	0.105	**	***	ns
SFA	51.56^{A}	48.65^{AB}	47.03^{AB}	39.72^{B}	51.46^{A}	45.49^{AB}	1.251	ns	ns	*
MUFA	30.46	39.65	40.37	37.10	36.26	36.45	0.971	ns	ns	ns
PUFA	12.13^{BC}	9.04^{D}	10.36^{CD}	15.27^{A}	11.16^{CD}	14.43^{AB}	0.338	***	**	ns
PUFA/SFA	0.24^{BC}	0.19^{C}	0.22^{BC}	0.39^{A}	0.22^{BC}	0.32^{AB}	0.014	**	*	ns
n-6	9.83^{AB}	8.28^{B}	8.47^{B}	12.11^{A}	10.17^{AB}	11.63^{A}	0.326	**	ns	ns
n-3	1.17^{BC}	0.75^{C}	1.45^{B}	2.83^{A}	0.67^{C}	2.55^{A}	0.400	***	***	**
n-6 /n-3	4.52^{BC}	11.74^{A}	3.33^{C}	3.42^{C}	12.61^{A}	3.73^{C}	0.941	ns	***	ns

$^{A, B, C, D}$ Means followed by a different letter within the same row are significant difference: *P<0.05; **P<0.01; ***P<0.001; ns: not significant difference (P>0.05) and SEM: standard error of mean, $^{\beta}$ S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$ F = effect of forage form (Fresh and Silage), SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C17:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + CLA + C18:3n3 + C20:3n6 + C20:4n6 + C22:2 + C22:6n3, CLA = Conjugated linoleic acid = *cis-9, trans-11*CLA + *trans-10, cis-12*CLA.

Table 5.7 Fatty acid profiles (g/100 g total fat) of peritoneal fat of the meat goats fed on different forage and regimes.

Item	Mulato II grass			Verano stylo			SEM	S^{β}	F^{γ}	S × R
	cut-and-carry	silage	grazing	cut-and-carry	silage	grazing				
C12:0	0.54	0.54	0.56	0.57	0.65	0.48	0.056	ns	ns	ns
C14:0	4.71^{AB}	4.81^{AB}	2.92^{B}	3.65^{AB}	5.13^{A}	3.94^{AB}	0.226	ns	*	ns
C15:0	0.53^{B}	0.43^{B}	0.83^{A}	0.39^{B}	0.36^{B}	0.43^{B}	0.024	**	*	ns
C16:0	26.21	26.35	23.87	24.71	26.33	23.50	0.465	ns	ns	ns
C16:1	1.53	1.43	1.65	1.32	1.27	1.12	0.062	ns	ns	ns
C17:0	1.31^{AB}	0.48^{C}	1.52^{A}	1.41^{A}	0.61^{C}	0.76^{BC}	0.070	ns	**	ns
C17:1	1.61	1.64	1.55	1.45	1.57	1.48	0.043	ns	ns	ns
C18:0	29.47	29.42	28.99	29.27	29.13	29.57	0.236	ns	ns	ns
C18:1n9	29.67	30.09	29.27	28.92	29.25	28.69	0.267	ns	ns	ns
C18:2n6	2.01^{B}	1.15^{C}	2.57^{AB}	2.66^{AB}	2.53^{AB}	3.03^{A}	0.106	**	*	ns
CLA	0.61^{AB}	0.40^{AB}	0.69^{A}	0.38^{AB}	0.35^{B}	0.44^{AB}	0.037	*	ns	ns
C18:3n3	0.74	0.68	0.66	0.46	0.32	0.65	0.049	ns	ns	ns
C20:3n6	0.06^{B}	0.08^{B}	0.22^{AB}	0.06^{B}	0.07^{B}	0.33^{A}	0.027	ns	*	ns
C20:4n6	0.03	0.06	0.34	0.07	0.07	0.25	0.040	ns	ns	ns
C22:2	0.05	0.08	0.31	0.34	0.02	0.16	0.051	ns	ns	ns
C22:6n3	0.05^{B}	0.07^{B}	0.48^{A}	0.06^{B}	0.05^{B}	0.38^{A}	0.038	ns	**	ns
SFA	62.76	62.04	58.68	60.01	62.21	58.66	0.608	ns	ns	ns
MUFA	31.69	32.08	31.29	32.80	33.16	32.47	0.290	ns	ns	ns
PUFA	2.93^{AB}	2.11^{B}	4.57^{A}	3.65^{AB}	3.06^{AB}	4.79^{A}	0.227	ns	*	ns

$^{A, B, C}$ Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001; ns: not significantly different (P>0.05) and SEM: standard error of mean, $^{\beta}$S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$F = effect of forage form (Fresh and Silage), SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C17:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + CLA + C18:3n3 + C20:3n6 + C20:4n6 + C22:2 + C22:6n3, CLA = Conjugated linoleic acid = *cis-9, trans-11*CLA + *trans-10, cis-12*CLA.

Table 5.7 (Cont.) Fatty acid profiles (g/100 g total fat) of peritoneal fat of the meat goats fed on different forage and regimes.

Item	Mulato II grass			Verano stylo			SEM	S^{β}	F^{γ}	S × R
	cut-and-carry	silage	grazing	cut-and-carry	silage	grazing				
PUFA/SFA	0.06^{AB}	0.05^{AB}	0.09^{A}	0.05^{AB}	0.03^{B}	0.08^{A}	0.005	ns	*	ns
n-6	2.09^{BC}	1.28^{C}	3.03^{AB}	2.80^{AB}	2.67^{AB}	3.61^{A}	0.132	**	**	ns
n-3	0.78^{ABC}	0.75^{ABC}	1.14^{A}	0.52^{BC}	0.37^{C}	1.23^{AB}	0.062	ns	*	ns
n-6 /n-3	2.80^{B}	1.72^{B}	2.73^{B}	6.38^{A}	6.66^{A}	3.52^{AB}	0.420	**	ns	ns

$^{A, B, C}$ Means followed by a different letter within the same row are significantly different: *P<0.05; **P<0.01; ***P<0.001; ns: not significantly different (P>0.05) and SEM: standard error of mean, $^{\beta}$S = effect of forage sources (Mulato II grass and Verano stylo), $^{\gamma}$F = effect of forage form (Fresh and Silage), SFA: saturated fatty acid = C12:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C22:0 + C24:0, MUFA: monounsaturated fatty acid = C16:1 + C17:1 + C18:1n9 and PUFA: polyunsaturated fatty acid = C18:2n6 + CLA + C18:3n3 + C20:3n6 + C20:4n6 + C22:2 + C22:6n3, CLA = Conjugated linoleic acid = *cis-9, trans-11*CLA + *trans-10, cis-12*CLA.

5.6 Discussions

The chemical composition (DM, NDF, ADF, EE and Ash) of Mulato II grass and Verano stylo, as offered to the experimental goats as cut-and-carry, silage and grazing were close to each other. This would imply that silage making with adding FJLB could preserve quality which was in agreement with earlier reports (Bureenok et al., 2006; Bureenok et al., 2011; Han et al., 2012). For proportion of FAs, the SFA, MUFA and PUFA contents in the fresh forage and forage silage were close to those reported in the Experiment I-I, I-II and II. The PUFA was major proportion in all experimental treatments. The C18:2n6 and C18:3n3 was the main composition. The SFA and MUFA were mainly come from C16:0 and C18:1n9, respectively. In general, ensilage by adding FJLB as additive would preserve quality of studied forages.

The body weight change indicated growth rate, which was high for the goats offered by Mulato II grass as cut-and-carry and for the goats raised by grazing Mulato II grass and Verano stylo. The higher growth rate would be mainly caused by higher intake (g/day) of the forages in agreement with the studies earlier (Goetsch et al., 2011b; Pralomkarn et al., 1995; Sahlu et al., 2004). When considered the digestibility of the goats, the digestibilies of crucial nutrients (OM, CP, ADF and NDF) for the goats were not different among all experimental groups. This implies that the growth rate of the goats was mainly positive relation to the total intake of the goats. Additionally, high digestibility of fat (EE) might be partly explained for better growth rate as low fat digestibility for the goats fed by silage of Mulato II grass and Verano stylo. However, there was a strange that the goats fed by Verano stylo as cut-and-carry had low growth rate and higher values of nutrient digestibilities when compared with the goats raised by grazing Verano stylo. The explanation for this discrepancy would

be that the goats fed on cut-and-carry had less choice for selecting parts of Verano stylo offered. This might result in lower feed intake with lower quality parts of Verano stylo and the goats tried to increase digestibility of nutrients. These would also be supported by the reports earlier that goats had higher total tract nutrient digestibilities of all-forage diets than sheep (Sales et al., 2012) and had capacity of physiological adaptation to various agro-climatic conditions (Morand-Fehr, 2005). In the meantime, when considered the goats fed the same regime of cut-and-carry with Verano stylo and Mulato II grass, there was rather different for growth rate of the goats. The explanation for this disagreement would be that Verano stylo had less resistance to dry season when compared with Mulato II grass (Hare et al., 2003).

The BUN concentration in plasma was in the normal range (13-26 mg/dl) reported in Thailand (Rattana et al., 2011), except for the BUN at 4 hours post feeding (28 mg/dl). This implies that all experimental treatments could keep BUN in the normal condition. The NH_3-N concentrations in rumen of the goats fed by Verano stylo as all studied regimes were low at the start of feeding when compared with those fed by Mulato II grass, but there were not different each other after feeding. It is presumed that Verano stylo containing high crude protein could make faster and more degradation of protein after feeding and then be absorbed into blood circulation. Thus, the current study found high BUN in plasma and low concentration of NH_3-N in rumen, which might be explained by the reasons mentioned earlier (Anbarasu et al., 2002; Chanjula et al., 2007; Darlis et al., 2000). The rumen pH of the goats in all experimental treatments dramatically reduced at 2 hours after obtaining feed, which would relate to the increase of acetic acid concentration in rumen at the same time after receiving diet. This explanation might be applied to the trend of negatively

relation between rumen pH and total VFA, mainly composed of acetic acids, at 4 hours after receiving diet. Generally, the results of this study for rumen pH and concentration of NH_3-N, AA, PA, BA and total VFA were in the similar trend of dynamic changes as reported earlier (Anbarasu et al., 2002; Darlis et al., 2000). Therefore, both Mulato II grass and Verano stylo offered to the goats as all three studied regimes had no different effect on rumen characteristics.

Carcass traits were influenced by the diet, genotype and their interaction (Casey and Webb; Dhanda et al., 1999a; Dhanda et al., 2003; Ryan et al., 2007). The current study should minimal difference for body component as percentage of slaughter weight. Dressing percentage (based on slaughter weight) in this study ranged from 37.5 to 45.1%, which a bit lower than the report of Dhanda et al. (1999) who found 41.3-45.1% dressing percentage for crossbred Boer goats while the other reports (Dhanda et al., 2003; Ryan et al., 2007) found higher values (51.0-54.0% and 41.8-51.3%, respectively) of dressing percentage. This discrepancy would be the results of difference genetic line of crossbred Boer goats or/and obtaining diets. There was evidence that grazing pasture of goat kids has no significant difference between genetic for live and carcass weight while goat kids fed the high concentrate diet have considerably greater (Goetsch et al., 2011a). The goats in the current study, Boer × Anglo-Nubian breed, were offered mainly forages with constant rate of concentrate and had lower BW than those reports. Therefore, the goats raised by grazing both Mulato II grass and Verano stylo had a trend of lower dressing percentage. It would indicate more influence of concentrate on the goats raised in pens. In the present study, the ranges of hot carcass weight, cold carcass weight, carcass length and loin eye area were 6.0 to 8.2 kg, 5.5 to 7.8 kg, 53.7 to 122 cm and 4.3 to 8.4 cm^2, respectively.

The hot and carcass weight, and loin eye area of the experimental goats was closed to the values in young meat goats reported earlier (Dhanda et al., 1999a), but the hot and cold carcass weight of older goats in the other reports (Dhanda et al., 1999a; Dhanda et al., 2003; Ryan et al., 2007) had higher values than those found in the current study. Thus, BW weight of goats at the slaughtering would be a major factor for determining hot and cold carcass weight and loin eye area. For the values of carcass length of the goats, there were two reports with large difference of carcass length, 51.5-54.6 cm^2 (Dhanda et al., 2003) vs. 96.5-99.6 cm^2 (Ryan et al., 2007), although the live BW of the goats in those reports were in almost the same range (25.5-27.2 kg vs. 25.0-31.1 kg) and the crossbred Boer goats were used in those two reports. This discrepancy would be hard to explain and also was found in the current study as high in the goats raised by grazing both Mulato II grass and Verano stylo but rather low in other treatments. However, the carcass length in the current study was deemed to positively relate with live and slaughter weight. The goats fed with silage for both Mulato II grass and Verano stylo in the present study were seemed to have low carcass performance. For lean percentage, the goats had a positively relation for % lean with hot carcass weight, carcass length and slaughter weight. Thus, feeding goats with Mulato II grass as cut-and-carry and as grazing Mulato II grass and Verano stylo give the good growth performance and carcass traits, with more preference for intake.

The meat $pH_{ultimate}$ of the experimental goats ranged from 5.40 to 6.11 and were within the acceptable range of $pH_{ultimate}$ (Webb et al., 2005; Yami and Merkel, 2008). High $pH_{ultimate}$ often occurs amongst temperamental animals, such as goats and generally highly prone to stress (Webb et al., 2005). A high $pH_{ultimate}$ of goat meat reflects depression of muscle glycogen as the results of stress or other factors (Dhanda

et al., 2003; Xazela et al., 2012), such as pre-slaughtering handling. The earlier report (Muir et al., 1998) indicated that grass-fed steers had higher $pH_{ultimate}$ values than grain-fed steers, which explained by more susceptible to pre-slaughtering from less acclimation to penning and handling for grass-fed steers. The meat $pH_{ultimate}$ of the goats raised by grazing both Mulato II grass and Verano stylo had low $pH_{ultimate}$ reflects high glycogen content and less stress to pre-slaughtering. These results would be explained that the goats raised by grazing had more glycogen content in muscle as more lean content in body and the goats kept in pen during nighttime resulting less stress to penning. In addition, there was a report (Mancini and Hunt, 2005) found that forage-based diet might promote oxidative metabolism, rather than anaerobic muscle metabolism and increase glycogen storage. The drip loss percentage of the goats raised by grazing Mulato II grass and Verano stylo had a high drip loss, which related to low $pH_{ultimate}$ of goat meat. This was in agreement with the report of Dhanda et al. (2002) who found that increasing pH could decrease cooking loss. For meat color, there were no difference for the values of L*, a* and b* at 45 minutes and 24 hours after slaughtering, except for b* (yellowness) at 24 hours post slaughtering as a trend of higher yellowness of goat meat fed silages. The increase of yellowness was in agreement with earlier report (Varela et al., 2004). There were the reports of yellow fat color of ruminants, which mainly resulted from carotenoid in forage (Ripoll et al., 2008; Ripoll et al., 2012; Röhrle et al., 2011; Varela et al., 2004). Thus, the increase of yellowness of goat meat in present study would be partly explained by carotenoid content in intramuscular fat. The shear force values were not affected by experiment treatments. However, all shear force values in the current study are fall in the

acceptable range of tenderness values (<6.0) and much lower than the reports earlier (Webb et al., 2005)

Chemical composition of goat meat (*longissimus dorsi*) in the current study was close to those in the reports earlier (Dhanda et al., 1999b; Dhanda et al., 2003), except for lower fat content in meat. This lower fat proportion might be the results of different genotypes and feed quality received. The major proportions of FAs in *longissimus dorsi* muscle of the studied goats were C16:0, C18:0, C18:1n9 and C18:2n6 while C18:3n3 and CLA were small proportion. This FA profile is quite difference from FA composition in diet, which contained mainly C16:0, C18:1n-9, C18:2n6 and C18:3n3 in accordance with earlier report (Rhee et al., 2000). The FA profile changes were the results of hydrogenation by microorganism in rumen of the goats as broadly discussed in the Experiment II. Briefly, microorganism in rumen added hydrogen bond to unsaturated FAs, such as C18:2n6 and C18:3n3, resulting in more proportion of SFAs, mainly C18:0 (Buccioni et al., 2012; Lourenço et al., 2010). The content of CLA was lowest in the goats fed by silage of Mulato II grass and Verano stylo, but the goats in other treatment had closed values of CLA proportion to the report earlier (Talpur et al., 2008). This implied that both silages might be not appropriate form for meat goats in order to elevate CLA level in meat. For the proportion of C18:3n3 was rather low when compared with the earlier reports (Dhanda et al., 2003; Talpur et al., 2008; Zervas and Tsiplakou, 2011). Hence, awareness of low CLA and C18:3n3 for forages fed as cut-and-carry, silage and razing would be paid more attention.

The ratio of PUFA/SFA in goat meat of this study ranged from 0.19 to 0.39, which is low the recommended ratio value of 0.45 for consumers (Enser et al., 1998).

However, the ratios of PUFA/SFA were in the closed range of the ratio in the reports earlier (Banskalieva et al., 2000; Talpur et al., 2008; Varela et al., 2004), but higher than the ratios reported by Dhanda et al. (1999b) and Webb et al. (2005) who found the ratios of PUFA/SFA ranged from 0.03 to 0.08 and from 0.06 to 0.08 (as re-calculation). This discrepancy would be explained by the influences of diets rather than phenotype. Therefore, feeding goats with forages and concentrate could increase PUFA and SFA ratio, but not reach recommended level. The n-6:n-3 ratios were extremely high for the *longissimus dorsi* muscle of the goats fed by both Mulato II grass silage (11.74) and Verano stylo silage (12.61) while the goats in the other groups had values of the n-6:n-3 at the range of 3.33 to 4.52. The ratio of n-6:n-3 at less than 4 was recommended for consumers. Thus, Mulato II grass and Verano stylo silages were not a good roughage source for decreasing the ratio and improving quality of meats. In the meantime, feeding goats by Mulato II grass and Verano stylo as cut-and-carry and grazing would result in improving quality of goat meats due to the n-6:n-3 ratio are much more affected by feeding than by genetics of the animal (De Smet et al., 2004). For FA profile of peritoneal fat, the main FA contents in the fat were C16:0, C18:0 and C18:1n9, which were closed to these fatty contents in *longissimus dorsi* muscle. The C18:2n6 in peritoneal fat was lower proportion than that in the muscle. The C18:3n3 were rather low when compared with the contents in meats and other results of the other researchers (Dhanda et al., 1999b; Dhanda et al., 2003), which studied in inter-muscular fat. As low values of sum PUFA, the ratio of PUFA/SFA was then rather low. The values of n-3 FAs were also low, leading to high variation of the n-6:n-3 ratio. There is not much information for FAs pattern in fat reported, then hardly to

interpretation. Therefore, it is possible that the FA profile in tissues might be depended on site of fat depot.

On overview of the current study, the meat goats were fed by Mulato II grass and Verano stylo with three feeding regimes, cut-and-carry, silage and grazing. The chemical compositions of forages were rather similar to each other, indicating the FJLB used for silage making could preserve forages. The goats with high intake of forages had high growth rate for the goats fed Mulato II grass as cut-and-carry and grazing, and Verano stylo as grazing, without nutrient digestibility difference. Rumen characteristics and BUN concentration in plasma of the goats in the present study were not affected by forage sources and feeding regimes. The experimental treatments had a tiny influence on microorganism population in rumen. For carcass traits, high lean percentage related to high hot and cold carcass weight, carcass length, loin eye area and feed intake. Meats of the goats raised by grazing both Mulato II grass and Verano stylo had low $pH_{ultimate}$, resulting in high drip loss and acceptable tenderness. Forage feeding would lead to more yellowness of *longissimus dorsi* muscle. The chemical composition of the goat meats was normal with low fat content. The majority of FA proportion in the goat meats was C16:0, C18:0, C18:1n-6 and C18:2n6, whereas low contents of CLA and C18:3n3. There was low ratio of PUFA/SFA for all goats, but the ratios of n-6:n-3 FAs were high for fed-forage silage goats and low for both forages fed by cut-and-carry and grazing. In the meantime, the main proportion of FAs in peritoneal fat was C16:0, C18:0 and C18:1n9, but rather low for the contents of C18:2n6, C18:3n3 and CLA. The ratios of PUFA/SFA and n-6:n-3 FAs in peritoneal fat were varied. Therefore, forage silages of Mulato II grass and Verano stylo prepared by adding FJLB would preserve quality of the silages and be used during shortage

period of roughage, but low growth rate, low lean percentage and high n-6:n-3 FAs should be given awareness. Grazing and cut-and-carry feeding of Mulato II grass and Verano stylo would be appropriate regime for the meat goats, except for fed-Verano stylo as cut-and-carry may result in low growth rate and lean production

5.7 Conclusion

Two forage sources, Mulato II grass and Verano stylo, with three feeding regimes, cut-and-carry, silage (prepared by adding fermented juice of epiphytic lactic acid bacteria; FJLB) and grazing could be used as forage feeding to the crossbred Boer (Boer × Anglo-Nubian), without effect on concentration of BUN in plasma and rumen fermentation. Cut-and-carry and grazing feeding regime for Mulato II grass and grazing Verano stylo could be applied to offer forages to meat goats with good responds of the meat goats; high growth rate, high lean yield, acceptable pH and tenderness and low fat contents with appropriate ratio of n-6:n-3 in *longissimus dorsi* muscle. However, the weaknesses of silages of both forages were low growth rate, low lean yield and high n-6:n-3 ratio in the meats and the Verano stylo offered as cut-and-carry gave low growth rate and lean yield of the meat goats.

5.8 References

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CHAPTER VI

GENERAL DISCUSSION AND IMPLICATION

Hitherto, food quality and safety have been increased as major concern of consumers. Meanwhile, food constraint and food security are now become more important in animal production as the increase of human population and drought period. Consequently, improving efficiency of animal production is a crucial strategy to solve these problems. Goats have been a source of human nutrition (Webb et al., 2005) and meat of goats could become an ideal choice of red meat for health conscious consumers (Carlucci et al., 1998; Johnson and Chen, 1995); due to its lower fat percentage compared to beef and lamb (Mahgoub et al., 2002) and a good source of desirable fatty acids (Banskalieva et al., 2000). Although concentrate was often provided important nutrients for goats, roughage is crucial source for goats and potential to improve productivity of meat goats. Because high quality diets with high level of concentrate is not affected the performance of goats, reflecting different responds of goats compared to cattle and sheep (Mushi et al., 2009; Ryan et al., 2007). Additionally, forage contains high PUFA (mainly C18:2n6 and C18:3n3) and has influences on goats by increasing the concentration of meat n-3 PUFA, compared with grain feeding (Aurousseau et al., 2004; Gatellier et al., 2004). However, the hydrogenating effect of the rumen microorganisms is an important factor to reduce PUFA proportion to finally deposit in ruminant meats (Wood et al., 2004).

The studies in this thesis have been performed to get more information and implication of using different forage sources and feeding regimes for meat goats. The first study was done in order to select appropriate forage source and silage preparation for meat goats. The first study composed of 2 phases. The first part of the study has shown that the experimental grasses (Purple Guineagrass, Mulato II grass and Napiergrass) and legumes (Thapra Stylo, Verano Stylo and Cavalcade), which were harvested at 30, 45 and 60 days after regrowth, would increase DM yield and content of DM, NDF, ADF and ash, but decrease OM, CP and EE with advancing maturity of the forage. This finding would be attributed to dilution effects of growth and increased concentrations of other metabolites, such as cellulose, hemicellulose and lignin (Clapham et al., 2005) with advancing age of forage. Almost FAs of the forages at 45 days after regrowth were in between that of the forage at 30 and 60 days after regrowth, which might be attributed by dilution effects of forage growth and structural constituents. The Mulato II grass and Verano stylo with the harvesting at 45 days after regrowth would contain proper chemical composition and properties including proper FA profile for ruminants. The second part of the first study found effects of different silage additives (molasses, cassava chips and FJLB) that grass silages in the present study had properties in the normal range of the criteria for well preserved silage as characterized as low pH (<5), low ammonia N (<90 g/kg of total N) and low concentration of butyric acid (<5.5 g/kg DM) (Phiri et al., 2007) while the legume silages at some intervals of maturity and/or some silage additives were not meet the criteria, but the silage of Verano stylo harvested at 45 days after regrowth, adding the FJLB had met the acceptable ranges of the criteria mentioned before. Therefore, adding FJLB into Mulato II grass and Verano stylo for ensiling would be possible to use in practice. The FA profiles of Mulato II grass silages composed main FAs; C16:0 (15.62-22.86%),

C18:0 (0.54-2.24%), C18:1n9 (1.67-3.97%), C18:2n6 (13.12-17.23%) and C18:3n3 (41.57-51.42%), while the rest of the analyzed FAs were lower than 2% of total fat for all additive treatments. These were closed to the main FAs of the Verano stylo silages composed of C16:0 (14.79-22.77%), C18:0 (1.50-5.42%), C18:1n9 (1.27-4.66%), C18:2n6 (16.38-25.98%) and C18:3n3 (35.00-45.02%). Thus, the studied additives would preserve FA contents of Mulato II grass and Verano stylo silages.

In the second study, ruminally fistulated meat goats were used to investigate effects of FJLB additive used for preparing silage of Mulato II grass and Verano stylo. The study has found that feeding silage of Mulato II grass and Verano stylo to meat goats had no difference effect on feed intake, rumen characteristics and FA pattern in rumen fluid, when compared with feeding the goats with fresh form of the forages. The microbial biohydrogenation presented in this study was in accordance with many reports earlier (Arvidsson et al., 2009; Buccioni et al., 2012; Lourenço et al., 2010; Woods and Fearon, 2009). From the results of the current study, silage made from Mulato II grass and Verano stylo with adding FJLB would be an alternative for preserving quality of the forages and could be used for roughage source for meat goats, especially during the shortage of fresh forage.

The third study, Mulato II grass and Verano stylo with three feeding regimes (cut-and-carry, silage and grazing) were applied to intact male meat goats to investigate these factors on production performance, rumen ecology, carcass and meat quality and FA profile of *longissimus dorsi* muscle and peritoneal fat. The results of the present study showed that goats with high intake of forages had high growth rate for the goats fed with Mulato II grass as cut-and-carry and grazing, and Verano stylo as grazing, without nutrient digestibility difference. Rumen characteristics and BUN concentration in plasma of goats in the present study were not affected by forage sources and feeding

regimes. The experimental treatments had a tiny influence on microorganism population in rumen. For carcass traits, high lean percentage related to high hot and cold carcass weight, carcass length, loin eye area and feed intake. Meats of the goats raised by grazing both Mulato II grass and Verano stylo had low $pH_{ultimate}$, resulting in high drip loss and acceptable tenderness. Forage feeding would lead to more yellowness of *longissimus dorsi* muscle. The majority of FA proportion in the goat meats was C16:0, C18:0, C18:1n-6 and C18:2n6, whereas low contents of CLA and C18:3n3. There was low ratio of PUFA/SFA for all goats, but the ratios of n-6:n-3 FAs were high (11.74 - 12.61) for fed-forage silage goats and low (3.33-4.52) for both forages fed by cut-and-carry and grazing. Hence, forage silages of Mulato II grass and Verano stylo prepared by adding FJLB would preserve quality of the silages and be used during shortage period of roughage, but low growth rate, low lean percentage and high n-6:n-3 FAs should be given awareness. Grazing and cut-and-carry feeding of Mulato II grass and Verano stylo would be appropriate regime for the meat goats, which be beneficial for health of consumers, except for fed-Verano stylo as cut-and-carry may result in low growth rate and lean production.

In conclusion, the results of this thesis would be implemented that raising meat goats by grazing Mulato II grass and Verano stylo would be the easier and cheap alternative for meat goat farmers in order to improve production performance, lean percentage and meat quality as higher unsaturated FAs and appropriate ratio of n-6 : n-3 FAs for consumers. In addition, the FJLB additive for ensiling from Mulato II and Verano Stylo would be practical for feeding meat goats, especially during constraint of fresh roughages, but feeding meat goats with silage prepared from Mulato II grass or Verano stylo would lower lean production and higher n-6:n-3 FA ratio in goat meat.

Thus, the further studies should emphasize on alleviating these undesirable results with cost benefits.

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BIOGRAPHY

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