Wageningen UR Livestock Research

Partner in livestock innovations

Effect of increased maturity of silage maize at harvest on conservation, dairy cow performance and methane emission

LIVESTOCK RESEARCH
WAGENINGEN UR

Abstract
Increasing maturity at harvest is evaluated as a cost-free method to increase starch concentration and rumen by-pass starch of maize silage, reducing methane emission. Results showed no effect on cow performance or silage stability, but the calculated effect on methane emission was low with the maize cultivar used.

Keywords
Dairy cow, maize silage, crop maturity, methane

Authors
R.L.G. Zom
A. Bannink
R.M.A. Goselink

Title
Effect of increased maturity of silage maize at harvest on conservation, dairy cow performance and methane emission

Report 578

LIVESTOCK RESEARCH
WAGENINGEN UR

Report 578

Effect of increased maturity of silage maize at harvest on conservation, dairy cow performance and methane emission

Projecttitel: Effectieve en kostenloze vermindering van de methaanuitstoot door de melkveehouderij met verder afgerijpte snijmaïs

R.L.G. Zom
A. Bannink
R.M.A. Goselink

March 2012

Preface

This project was funded by the Dutch Dairy Board and the Dutch Ministry of Economics, Agriculture and Innovation within the programme "Reduction Other Greenhouse Gases", which supports research to reduce greenhouse gases other than carbon dioxide. Methane is one of the most important greenhouse gases in dairy farming, and is mainly produced as a by-product of fermentation in the rumen. In the present project, the effects of delaying harvest of maize silage on the starch content, starch digestibility and related methane emissions are evaluated. In addition, some practical aspects of feeding maize silage of increased maturity on dairy farms are considered, such as silage preservation and stability as well as feed intake and milk production of cows.

R.L.G. Zom
A. Bannink
R.M.A. Goselink

Samenvatting

Inleiding

Snijmaïs is na gras het belangrijkste voedergewas in Nederland. Het is een belangrijke bron van energie en zetmeel in het rantsoen van melkvee. Door de vertering van zetmeel uit snijmaïs van de pens naar de darm te verschuiven zou de melkveehouderijsector theoretisch de methaanemissie met 5% (ca. 2,1 Mton) CO_2 equivalenten kunnen verminderen (Tamminga et al., 2007). Verschuiven van de zetmeelvertering van de pens naar de darm kan eenvoudig worden gerealiseerd door snijmaïs verder te laten afrijpen en bij een hoger drogestof (DS) gehalte te oogsten dan wat nu in de praktijk gebruikelijk is en geadviseerd wordt. De maatregel om snijmaïs ver te laten afrijpen is beoordeeld als eenvoudig met een hoge praktische toepasbaarheid (Tamminga et al., 2007). Bovendien heeft recent onderzoek laten zien dat het verder laten afrijpen ook een gunstig effect kan hebben op de zetmeel- en voederwaardeopbrengst van snijmaïs (van Schooten et al., 2006). Bij een hogere zetmeel- en voederwaardeopbrengst van verder afgerijpte snijmaïs zal de regionale zelfvoorziening toenemen waardoor de behoefte aan van elders geïmporteerd zetmeelrijk krachtvoer (bijv. granen) kleiner wordt, hetgeen de food-for-feed concurrentie vermindert. Het oogsten van verder afgerijpte snijmaïs vraagt geen investeringen en is daarom een kosteloze maatregel om de methaanmissie te beperken. Een bijkomend voordeel is dat van deze maatregel geen afwenteling van de broeikasgasemissie plaats zal vinden naar het buitenland of naar andere sectoren.

Ondanks de hoge potentie van deze maatregel om de methaanemissie te verminderen, wordt het overgrote deel van de snijmaïs geoogst bij een DS gehalte in de range van 30 tot 35% DS. Onder veehouders bestaat er namelijk onzekerheid ten aanzien van mogelijk negatieve effecten op de voeropname en een grotere broeigevoeligheid wanneer snijmaïs verder is afgerijpt. De acceptatie van de voorgestelde maatregel zal toenemen wanneer veehouders overtuigd worden van de mogelijkheid dat problemen die mogelijk ontstaan rondom de bewaring, conservering, en kwaliteit kunnen worden voorkomen en dat het verder laten afrijpen van snijmaïs geen nadelige effecten heeft op de voeropname en melkproductie.

Vraagstelling

Tot op heden ontbrak het aan duidelijke onderzoeksgegevens die inzicht geven op de effecten van het oogsten van verder afgerijpte snijmaïs bij een hoger DS-gehalte buiten de gangbare range van 30 tot 35% DS. Om deze leemte op te vullen is onderzoek uitgevoerd naar de voeropname en melkproductie van melkkoeien, in-situ pens- en darmverteerbaarheid, en broeigevoeligheid van snijmaïs geoogst bij 30% (MS30), 34% (MS34), 38% (MS38) en 42% (MS42) drogestof (DS). Voor een zo breed mogelijke praktische toepasbaarheid van de resultaten is het onderzoek uitgevoerd bij twee contrasterende krachtvoersamenstellingen, die representatief zijn voor de mogelijke variatie in krachtvoersamenstelling in de praktijk. Deze krachtvoeders hadden beide een gelijke energie (VEM) en eiwitinhoud (ruw eiwit, DVE) maar verschilden in het gehalte snel en langzaam afbreekbare koolhydraten (hoog gehalte langzaam afbreekbare koolhydraten LC, hoog gehalte snel afbreekbare koolhydraten HC).

Proefopzet

Het onderzoek is uitgevoerd in de vorm van een voederproef met 64 verse HF melkkoeien (waarvan 16 vaarzen en 48 oudere dieren). Deze melkkoeien werden verdeeld over 8 blokken van elk 8 dieren op basis van overeenkomsten in lactatienummer, verwachte afkalfdatum, lichaamsgewicht en melkproductie in de voorafgaande lactatie. De 8 dieren van elk blok werden *ad random* toegewezen aan 8 behandelingen. De 8 behandelingen bestonden uit 4 basisrantsoenen met snijmaïskuil geoogst bij 30, 34, 38 en 42% DS, die elk werden herhaald bij de twee krachtvoersoorten LC en HC. Krachtvoer LC en HC waren identiek voor energie-inhoud (1055 VEM/kg DS), darmverteerbaar eiwit (123 g DVE/kg DS) en de onbestendig eiwit balans (40 g OEB/kg DS). Krachtvoer LC bevatte 90 gram water oplosbare koolhydraten en 386 g neutral detergent fiber (NDF) per kg DS. Krachtvoer HC bevatte 119 g water oplosbare koolhydraten en 312 g NDF per kg DS. Het basisrantsoenen werd onbeperkt gevoerd met registratie van de individuele voeropname. Het rantsoen bestond op drogestof basis uit 61% snijmaïs, 28% graskuil, en 10% sojaschroot en 0.5% mineralen en zout. Daarnaast werd individueel 8.5 kg drogestof van één van beide krachtvoersoorten LC of HC verstrekt via krachtvoerautomaten.

Gedurende een 15 weken durende proefperiode werden de individuele voeropname, melkproductie, gewicht en body conditiescore (BCS) geregistreerd. De pensverteerbaarheid van de snijmaïssilages werd bepaald met behulp van de nylonzakjes methode met pensgefistuleerde melkkoeien. De

darmverteerbaarheid werd bepaald met behulp van de mobile nylon bag methode met koeien voorzien van canules in het duodenum (twaalfvingerige darm) en het einde van het ileum (dunne darm). Om verschillen in broeigevoeligheid te bepalen werd gedurende een periode van een maand dagelijks het temperatuursverloop onder de oppervlakte aan de open voorzijde van de maiskuilen gemeten. Tevens werd de dichtheid (kg ds/m^3) van de kuil bepaald door twee blokken uit te snijden. Gedurende één week is het dagelijkse temperatuursverloop in deze kuilblokken gemeten op ca. 1/3 (40 cm) en 2/3 (90 cm) van de hoogte van de kuil.

Resultaten
Het zetmeelgehalte van de snijmaiskuil nam toe met het DS-gehalte en bedroeg 381, 396, 415 en 433 g zetmeel/kg DS voor respectievelijk MS30, MS34, MS38 en MS42. Het NDF gehalte nam af met toenemende rijpheid van 366, 350, 345 en 342 g NDF/kg DS voor respectievelijk MS30, MS34, MS38 en MS42.

De toenemende rijpheid leidde niet tot verschillen in de drogestof, energie- en eiwitopname. Bij een toenemende rijpheid nam de zetmeelopname toe, terwijl de opname van NDF en ADF afnam (Table 4). Het afrijpingsstadium van de snijmaïs had desondanks een effect op de melkgift, melksamenstelling, gewicht en BCS (Table 5). De melkvetproductie was lager bij MS42 in vergelijking met MS34 en MS38 (p<0.05).

Bij het krachtvoer met een laag gehalte aan snel fermenteerbare koolhydraten waren de melkgift, eiwit- en lactoseproductie hoger dan bij het krachtvoer met het hoge gehalte aan snel fermenteerbare koolhydraten (P < 0.05). Er waren geen interacties tussen krachtvoersamenstelling en het afrijpingsstadium van snijmaïs. Hieruit kan worden geconcludeerd dat de effecten het afrijpingsstadium van snijmaïs op de dierprestaties veralgemeniseerd kunnen worden voor een breed spectrum van krachtvoersamenstellingen.

De verschillen in afrijpingsstadium zorgden voor kleine verschillen in de pens- en darm afbreekbaarheid van het zetmeel in de snijmaïskuilen (Table 6). Op basis van de in-situ nylonzakjes afbreekbaarheid van zetmeel kon worden berekend dat de hoeveelheid maiszetmeel die de pens passeert naar de darm toeneemt met het afrijpingsstadium. De hoeveelheid pensbestendig zetmeel bedroeg 1760, 1817, 1943 en 2110 g per dag voor respectievelijk MS30, MS34, MS38 en MS42. De gegevens van de mobile nylon bag afbraak geven aan dat praktisch alle pensbestendige zetmeel dat de darm binnenkomt ook daadwerkelijk in de darm wordt verteerd. De residuen bedroeg 1.47%, 1.58%, 1.77% en 1.57% voor respectievelijk MS30, MS34, MS38 en MS42. De berekende hoeveelheid zetmeel die in de dikke darm binnenkomt bedroeg slechts 26, 29, 34 en 33 g/dag voor respectievelijk snijmaïssilage MS30, MS34, MS38 en MS42.

Met een goed inkuilmanagement (een goede verdichting van de kuil, het gebruik van een gronddek en een voersnelheid van 0.15 m/dag (1 m per week)) is het mogelijk om de kuilen broeivrij te houden. De gemiddelde temperaturen onder het frontoppervlak van de maiskuilen waren 15.4, 17.2, 18.5 en 20.3˚C voor respectievelijk MS30, MS34, MS38 en MS42. Deze temperaturen waren vergelijkbaar met de omgevingstemperatuur. Metingen gedurende een week aan een uitgesneden blok lieten zien dat toename in temperatuur van uitgesneden kuilblokken vergelijkbaar was voor alle snijmaiskuilen. De dichtheden van de kuilen bedroeg 198, 208 286 en 255 kg DS per m^3 voor respectievelijk MS30, MS34, MS38 en MS42.

Op basis van modelberekeningen volgens de TIER 3 methode (Bannink et al., 2011) kan worden geconcludeerd dat de absolute methaanemissie (of als % van de GE-opname) afneemt met toenemende rijpheid van snijmaïs als gevolg van een verschuiving van de zetmeelvertering van de pens naar de darm. De verschillen in de reductie van methaanemissie als gevolg van toenemende rijpheid zijn geringer dan door Tamminga et al. (2007) op basis van literatuurgegevens werd gesuggereerd. Dit komt omdat het zetmeelgehalte in alle onderzochte snijmaiskuilen hoog was en daarnaast de verschillen in zetmeelgehalte en zetmeelbestendigheid klein waren. Ter vergelijking, de CVB tabel (CVB, 2007) geeft een waarde van 304 g zetmeel per kg drogestof voor snijmaiskuil met ca. 300 g drogestof per kg, hetgeen 25% lager is dan waargenomen in deze studie. Voor snijmaïs met meer dan 320-360 g drogestof geeft de CVB een waarde van 342 g zetmeel per kg drogestof. Dit is 17% lager dan snijmaïs met een vergelijkbaar drogestof gehalte (snijmaïs M34) in deze studie. De geringe verschillen in zetmeelgehalte kunnen een gevolg zijn van rassenveredeling. Het zetmeelgehalte is een belangrijk criterium voor toelating op de rassenlijst. Mogelijk heeft de selectiedruk er voor gezorgd dat het zetmeelgehalte in snijmaïs in recente snijmaisrassen hoger is dan wordt gesuggereerd op basis van de literatuur en de CVB tabel. Mogelijk spelen ook de relatief gunstige groeiomstandigheden een rol die kunnen bijgedragen aan een hoog zetmeelgehalte en geringe verschillen in zetmeelgehalte tussen oogsttijdstippen. Vanwege numerieke verschillen in de drogestof opname en de melkproductie was het effect van het oogststadium op de methaanemissie

minder groot dan verwacht; er waren geen verschillen in de berekende methaanemissie per kg FPCM of verschillen in methaanemissie tussen de afrijpingsstadia. (Table 8).

Conclusie

Uit deze studie kan worden geconcludeerd dat het oogsten van snijmaïs in een verder afgerijpt stadium, dat wil zeggen boven het gangbare traject 30-35% drogestof, leidt tot een hogere zetmeelopname, meer pensbestendig zetmeel en een grotere doorstroming van zetmeel naar de dunne darm. Op basis van darmverteerbaarheidsonderzoek met de mobile nylon bag methode kon worden vastgesteld dat vrijwel alle pensbestendige zetmeel werd afgebroken in de dunne darm, waardoor de zetmeeluitstroom naar de dikke darm of zelfs mest verwaarloosbaar klein zijn.

Oogsten van snijmaïs in een verder afgerijpt stadium heeft geen significant effect op de voeropname of melkproductie. Het later oogsten van snijmaïs bij een hoger drogestof gehalte is een adequate methode om een hoger zetmeelgehalte in het rantsoen en een hogere pensbestendigheid van het zetmeel te verkrijgen en daarmee een deel van de zetmeelvertering naar de darm te verschuiven. Door verschuiving van de zetmeelvertering naar de darm, neemt de methaanemissie per kg drogestof af.

Het later oogsten van snijmaïs bij een hoger droge stofgehalte had in de huidige proef geen nadelige gevolgen voor de voeropname, melkproductie of broeigevoeligheid van snijmaiskuilen. Bij goed inkuilmanagement (zoals een goede kuilverdichting, afdekken met zand en voldoende voersnelheid) kan deze methode zonder bezwaar in de praktijk worden toegepast en worden beschouwd als een kosteloze maatregel om de methaanemissie uit de melk veehouderij te verminderen. Het bestaande oogstadvies in het Handboek Snijmaïs om te oogsten bij maximaal 36% drogestof kan worden verruimd tot maximaal 42%.

Summary

In the Netherlands, after grass, maize silage is the most important fodder crop for ruminants. Maize silage has a relatively high energy content, and is a major source of starch in dairy cow rations. Increasing the starch content and the proportion of by-pass starch of maize silage in dairy cow rations has been suggested as an option to reduce the enteric methane emissions by 200,000 tons of CO2 equivalents. A higher proportion of starch and by-pass starch in the diet of dairy cows can be achieved by harvesting maize silage at a more mature stage (>350 g DM/kg in the whole crop). Increased maturity at harvest results in an increased grain filling and starch to NDF ratio, and an increased fraction of rumen by-pass starch. These changes can influence the microbial population in the rumen environment, the rate of digestion and the enteric emission of methane. Delaying harvest until the crop has reached a very mature stage could be a simple and low-cost measure to increase dietary starch, thereby reducing methane emissions from dairy farms. However, farmers are reluctant to apply this measure because there are many uncertainties about the effects of high-DM maize silage on feed intake and subsequent milk yield, and the risks of feed losses during the storage of silage due to heating and moulding.

This study evaluated the effect of silage maize ensiled at targeted dry matter (DM) contents of 300 (MS30), 340 (MS34), 380 (MS38) and 420 (MS42) g/kg fresh weight. Maize silage was fed to dairy cows in a combination with a high degradable carbohydrate (HC) or low degradable carbohydrate (LC) concentrate and effects were observed on nutrient intake, milk yield and milk composition, in-situ nylon bag degradation, silage preservation and emissions of methane were calculated. Sixty-four multiparous Holstein-Friesian dairy cows in their first week of lactation were assigned to the eight dietary treatments according to a randomized complete block design. The eight dietary treatments consisted of a factorial combination of the four maize silage and the two concentrates. Maize silages were offered ad libitum as part of a basal roughage mixture, while the concentrates were given at the rate of 8.5 kg DM/cow/day during the 15 weeks experimental period. The contents of starch in the maize silages increased (381 to 433 g/kg DM), and those of neutral detergent fibre (NDF) (366 to 341 g/kg DM) decreased with increasing maize harvest maturity. The intake of DM, crude protein and energy did not differ (P>0.05) across the maize silages. However, the intake of starch increased (P<0.01), and intake of NDF and ADF decreased with increasing maturation. Milk yield and composition were similar (P>0.05) across the maize silages. Yield (kg/d) of milk, milk protein and milk lactose was higher (P<0.05) on the LC compared to HC concentrate treatment. Maturity of maize silages at harvest did not affect the production performance of dairy cows. There were only marginal effects on the in-situ rumen degradability of the maize silages. Mobile nylon bag degradation indicated that rumen by-pass starch is almost entirely digested in the small intestine. Model simulations indicated that absolute emission of methane was (or as % of GE intake) reduced with increased harvest maturity. However, there were no effects of increasing harvest maturity on the enteric methane emissions per unit of fat and protein corrected milk. This is because the effects of increased maturity on starch concentration, starch to NDF ratio and the concentration of by-pass starch were less than expected. The effects of increasing harvest maturity on starch content were smaller than suggested by literature. In addition to that, at all stages of harvest maturity, the observed starch contents were much higher than reported elsewhere. Nevertheless, increasing maturity at harvest remains a good and price low manner to increase starch concentration and the proportion of rumen by-pass starch to reduce methane emission, but the calculated effects on methane emission are small. With appropriate silage management such as good compaction, using a sand load on top of the clamp and a sufficient rate of feed removal from the clamp (0.15 m/d) heating and moulding can be avoided.

Table of contents

1 Introduction

Silage maize is a major forage component in the ration of dairy cows, under most dietary regimes. The crop has a relatively stable yield, high energy content, good ensiling characteristics, and inclusion of maize silages in grass or grass silage based diets can increase feed intake, milk yield and milk protein content (Phipps et al., 1995; O'Mara et al., 1998; Phipps et al., 2000). As a result, like many other European countries, the area used for silage maize production in the Netherlands has increased from 5,000 ha in 1970 to 240,000 ha in 2004 (Schröder 1998; Barrière et al., 2006).

In the Netherlands, farmers are advised to harvest silage maize at a stage of maturation above 300 g dry matter (DM) per kg fresh weight (FW) and below 360 g DM/kg FW (Handboek Snijmais, www.handboeksnijmais.nl). Harvest above 300 g DM/kg FW is recommended to avoid effluent losses form the silage clamp. The upper value of 360 g DM/kg FW is a cut-off value. This value was chosen because there is little data available on the effects of dairy cow performance, dry matter intake (DMI), digestibility and losses during storage from maize silages harvested with a DM content above 350 g DM/kg FW. It is well-documented that an increased maturity at harvest results in an increased grain filling, starch-to-NDF ratio and increased fraction of rumen by-pass starch in maize silages (Bal et al., 2000; Phipps et al., 2000). An increased starch concentration and fraction of rumen by-pass starch can influence the rumen environment and digestion. Tamminga et al. (2007) postulated that the methane emission by dairy cows in the Netherlands could be reduced with 5% (or 2.1 Mton CO_2 equivalents) by feeding dairy cattle rations with higher concentrations of starch and a higher proportion of rumen by-pass starch. Therefore, inclusion of maize silage which is harvested at a more mature stage (>350 g DM/kg FW) in the diet of dairy cows could be an option to increase the supply of starch and rumen by-pass starch and thereby reducing the emission of methane by dairy cows. Moreover, a recent study indicated that increased harvest maturity of silage maize results in higher yields of dry matter (DMY), starch and energy (van Schooten et al., 2006). Therefore, increasing maturity may also contribute to an increased self-sufficiency rate for feed and hence reduce the farm imports of feeds and forage and the food-for-feed competition. In conclusion, increasing harvest maturity could be a simple and zero-cost measure to increase dietary starch with a larger proportion of rumen by-pass starch, thereby reducing methane emissions from dairy farms, without negative side effects on greenhouse gas emissions in other sectors or foreign countries.

However, increased rumen by-pass starch could be a risk for increased hindgut fermentation and losses of starch in manure. Because of the uncertainties about the effect of increasing harvest maturity of silage maize on DMI and subsequent milk yield; intestinal digestibility of starch and feed losses during the storage of silage due to heating and moulding, farmers are reluctant to apply this measure to reduce methane emission from dairy cows. Therefore a study was conducted to elucidate the effects of increased harvest maturity of silage maize on:
1) Feed intake and milk production by dairy cows
2) Ruminal and ileal digestibility of starch
3) Silage preservation
4) Effects on enteric methane production

Furthermore, the study included also the possible effects of concentrate composition and the interactions with the type of maize silage. In practice, a large variation exists in concentrate composition. Therefore, each maize silage treatment was combined with two different concentrates that were iso-nitrogenous and iso-energetic, but differed in carbohydrate composition. The potential effects of the increased maturity of maize silage at harvest were calculated using model simulation. The final aim is to develop practical nutritional strategies for dairy farmers to reduce the emission of methane by dairy cows fed maize silages.

Table 1   Abbreviations

Feed and diet composition	
FW	Fresh weight
DM	Dry matter
OM	Organic matter
CP	Crude protein (=N×6.25)
Cfat	Crude fat
WSC	Water soluble carbohydrates
NDF	Neutral Detergent Fiber
ADF	Acid Detergent Fiber
ADL	Acid Detergent Lignine
OMD	Organic Matter Digestibility
NE_L	Net Energy for Lactation (1 NE_L = 6.9 kJ)
DVE	Intestinal digestible protein (Darm Verteerbaar Eiwit)
OEB	Rumen degradable protein balance (Onbestendig Eiwit Balans)
Cow performance	
FPCM	Fat and protein corrected milk
	FPCM = Milk yield (kg) × (0.337 + 0.116 × Milkfat%+ 0.06 × Milkprotein%)
BCS	Body condition score
In-situ degradation	
W	Washable fraction
D	Potential in-situ degradable fraction
U	In situ undegradable fraction
kd	In situ degradation rate
kp	In situ passage rate

2 Materials and methods

2.1 Feeding trial

2.1.1 Silages

Maize silages were prepared from a single crop (cv. Atrium; Force Limagrain Nederland BV, Rilland, the Netherlands), sown on clay soil on 20 April 2009 at a density of 100,000 seeds/ ha (10 plants/m^2) and row spacing of 0.75 m, at the Waiboerhoeve experimental farm of Wageningen University and Research Center, Lelystad, The Netherlands (52° 5 N and 5° 5 E). The crop was fertilised with 50 tons of cattle slurry per ha (containing 4 kg N/ton and 1.3 kg P_2O_5/ton), and an additional 30 kg N/ha and 30 kg P_2O_5/ha as ammonium phosphate. The maize was harvested and ensiled at target DM contents of 300 (MS30), 340 (MS34), 380 (MS38) and 420 (MS42) g/kg FW. To determine the targeted harvest DM, 5 plants from 5 randomly selected spots in each cross section of each plot were sampled twice weekly, chopped, and dried in an oven at 103°C for 24 h. The frequency was increased to daily sampling when the difference with the target DM content was less than 30 g/kg. The actual DM content of the crop was close to the targeted DM contents (Table 2). All silages were made with the same precision chop harvester (John Deere 7750) using the identical machine settings. Theoretical length of cut was 6 mm and roll-clearance of the kernel processor was 1 mm, to ensure that all kernels were sufficiently crushed. The maize silages were stored in bunker silos and compacted with a heavy weight tractor and a wheel loader. Additives to improve the ensiling process were not used. The silages were sealed airtight with two layers of 0.15 mm polyethylene plastic sheets and covered with a 20 cm thick sand load. The total silage clamp was covered with a protection sheet held down with sand bags.

The grass silage was prepared from first-cut perennial ryegrass (*Lolium perenne L.*) cultivars (BG3; Barenbrug, Oosterhout, the Netherlands), mowed on 1 May 2009 with a disc mower and conditioner. The grass was wilted for 36 h with 20 h of sun, and tedded twice in the field. The average temperature of the day was 20.4°C and average temperature of the night was 7°C. Grass was ensiled in bunker silos, compacted and sealed as described for the maize silages.

Table 2 Harvest dates and weather conditions during the preceding period after flowering

Date	Harvest parameter				Weather after flowering			
	DAF[1]	DAS[2]	DM[3]	Yield (ton DM/ha)	Mean T (°C)	Max T (°C)	Min T (°C)	T_{sum}[4] (°C*d)
14-09-2009	64	146	296	16.28	17.6	22.7	12.5	500
23-09-2009	73	155	341	17.28	15.1	20.0	10.0	546
05-10-2009	85	167	396	17.02	12.4	16.7	8.1	574
14-10-2009	94	176	421	17.08	10.8	15.0	6.6	581

[1] Days after flowering
[2] Day after sowing
[3] Crop dry matter content at harvest (n=10)
[4] Temperature sum (°C*d, with a base temperature of 10° C; Sibma et al., 1987) after flowering

2.1.2 Experimental design, animals and diets

Sixty-four multiparous Holstein-Friesian dairy cows were assigned to eight blocks, based on similarity in parity, milk yield during the previous lactation, body weight and DM intake. The eight cows from each block were randomly distributed over eight dietary treatments (n = 8 cows per dietary treatment), creating a randomized block design. The eight dietary treatments consisted of a factorial combination of the four maize silages (MS30, MS34, MS38 and MS42) and two types of concentrate: a high degradable carbohydrate (HC; low NDF, high water soluble carbohydrates (WSC)) and low degradable carbohydrate (LC; high NDF and low WSC) concentrate. Cows were adapted to the experimental diets and feeding regimes just after calving and data was collected from the second week after calving until 17 weeks after calving (30 March to 27 August 2010).

The four maize silages were offered *ad libitum* as a part of the basal roughage mixture, containing 61% maize silage, 28% grass silage, 10% soybean meal, 0.45% mineral mixture (190 g Ca, 45 g Na, 1200 mg Cu, 2500 mg Zn, 3000 mg Mn, 120 mg I and 34 mg Se on DM basis) and 0.34% salt (NaCl;

380 g Na and 570 g Cl on DM basis) on a DM basis. The concentrates were given at a rate of 8.5 kg DM/cow/day. Fresh mixed roughage diets were prepared daily using a self-propelled mixer equipped with a cutter loader system and an electronic weighing unit. The roughage mixtures were fed in individual weighing troughs containing data loggers, which recorded the roughage intake after each visit for individual cows. The troughs were continuously accessible except during the milking period. The concentrates were fed individually using three transponder-controlled concentrate dispensers. The concentrates were dispensed at a rate of 0.3 kg/min. The total daily allowance of the concentrates was partitioned over 6 consecutive time windows of 4 hours each. The eight dietary treatments were formulated to be iso-nitrogenous and iso-energetic and only differed in maize silage maturity and carbohydrate degradability of the concentrates. The nutrient requirements of the cows were calculated according to CVB (2007) formulas.

2.1.3 Data recording and sampling

Fresh roughage mixtures were sampled daily, the individual ingredients of the roughage mixtures were sampled twice a week and concentrates once after delivery of a new batch for DM analysis. For chemical and FA analysis, samples of individual feedstuffs and the roughage mixture were taken weekly and frozen immediately at -20° C. The DM intake and milk yield of individual cows was recorded daily throughout the experiment with cows being milked twice daily at 06:00 and 18:00 h. Milk samples were taken weekly from four consecutive milkings. The morning and evening milk samples were pooled (1:1 ratio) separately to two composite milk samples. The samples were stored at 4°C until analysed for fat, protein, lactose and milk somatic cell count. To measure changes in body weight, the pre-calving body weights were recorded weekly whereas post-calving body weights were recorded automatically twice a day at the entrance of the milking parlour. The body condition score of each cow was recorded weekly by an experienced observer on a scale from 1 (thin) to 5 (fat) with 0.25 intervals (Edmonson et al., 1989).

2.2 In-situ ruminal and intestinal mobile nylon bag degradability

The in situ nylon bag ruminal degradation of the maize silages was determined at the Waiboerhoeve, Lelystad, the Netherlands using 3 rumen fistulated mid-lactation dairy cows that were fed a ration consisting of maize silage, grass silage and compound concentrate. From each maize silage, samples were taken 3 months after ensiling. Disappearance of OM, starch, NDF and N from samples of each maize silage (5 g DM) in nylon bags was determined after 0, 2, 4, 8, 16, 32, 72 and 336 h of rumen incubation according to the CVB protocol (CVB, 2003). The washout fraction (W-fraction) was defined as the fraction disappearing from the 0 h nylon bag incubation by washing with cold tap water in a washing machine for 45 min. After incubation, the nylon bags were pooled by incubation time and analysed for dry matter, starch and NDF content. The data were fitted on the model of Ørskov & McDonald (1979):

$$Y(t) = W + D \times (1 - e^{kd \times t})$$

In which:
- Y is the fraction disappeared from the nylon bag at time t (h);
- W is the washable fraction (%);
- D is the non-washable potentially degradable fraction (%) calculated by D = 100-W-U, where U is the undegradable residue after 336 h rumen incubation;
- kd is the degradation rate (%/h);
- t is time (h).

The proportion of rumen by-pass starch (B) was calculated as:

$$B = (kp/(kp+kd) \times D + 0.1 \times W$$

In which:
- kp is the rumen passage rate (%/h);
- kd is the degradation rate (%/h);
- D is the non-washable potentially degradable fraction (%);
- W is the washable fraction (%)

It was assumed that 10% of the washable starch can be considered as rumen by-pass starch. The in-situ nylon bag degradation data were used as input to the simulation model.

The ileal digestibility of the maize silages was determined by the mobile nylon bag technique at the Research Centre Foulum of the Danish Institute of Agricultural Sciences (DIAS) according to the procedures described by Norberg et al. (2007). The mobile nylon bags containing intact residues after a 12-h (CP and starch) or 24-h (NDF) ruminal incubation were pre-treated using a 2-step procedure (Volden and Harstad, 1995) to simulate abomasal digestion before insertion through the duodenal cannula. To assess the effect of hindgut fermentation on starch digestibility, approximately half of the bags were collected from the ileum and half from the faeces.

2.3 Silage preservation study

Daily, during the month of June, immediately before removal of silage from the clamps, ambient temperature and the temperature of the maize silage clamps were measured. Temperature of each silage clamp was measured on 6 spots; at 3 spots approximately at 1/3 of the height of the clamps (0.4 – 0.45 m) and at 3 spots at approximately 2/3 of the height of the height of the clamp (0.85-0.90 cm). At each spot a digital thermometer was inserted 30 cm in the front surface of the clamp.

In order to mimic heating of a silage clamp in a practical situation in which silage is removed from the clamp daily leaving the clamp open at the front site, one block (1.2 m height × 1.5 m width × 0.8 m depth) was cut loose from the clamp. The dimensions and the weight of the maize silage blocks were measured to calculate the density of the silage and each block was covered with 4 cm thick polystyrene foam insulation plates at three sides. During one week, the temperature of the block maize silage was measured daily using a digital thermometer that was inserted 30 cm in the silage blocks.

2.4 Chemical analysis

All feed samples were freeze-dried and ground to pass through a 1 mm screen, and analysed for DM, ash, crude protein (CP), crude fat (Cfat), NDF, acid detergent fibre (ADF), acid detergent lignin (ADL), starch and sugar content. DM content was determined by oven drying at 103°C for 24 h (ISO 6496; ISO, 1983), ash after incineration at 550°C (ISO 5984; ISO, 1978) and CP was determined using the Kjeldahl method (ISO, 5983; ISO 2005) for N and multiplication by 6.25. ADF and ADL was determined according to Van Soest (1973). NDF was analysed according to Van Soest et al. (1991) with some modification as described by Khan et al. (2009). Crude fat was determined using the Berntop method with pre-acid hydrolysis (ISO, 6492; ISO 1999). Sugars were determined as described by Van Vuuren (1993). The starch content was determined as glucose using the amyloglucosidase method (ISO, 5914; ISO 2004) after an initial extraction of the samples with 40% ethanol (to remove the sugar fraction). Ammonia was determined according to the Berthelot method as modified by Schneider (1976). The feeding value in vitro organic matter digestibility (OMD) was determined according to the method of Tilley and Terry (1963), net energy for lactation (NEL) was calculated according to Van Es (1978) and protein value was calculated according to the Dutch protein evaluation system as described by Tamminga et al. (1994) to determine DVE and OEB.

2.5 Model simulation

A dynamic, mechanistic model of enteric fermentation in dairy cows was used to estimate the effect of maize silage and type of concentrate, DMI on variation in methane (CH_4) emission from enteric fermentation in dairy cows according to the IPCC Tier 3 approach. The model and simulations procedures are extensively described by Bannink et al. (2011). The inputs for the model were data obtained from the feeding trial (DMI, chemical analyses of the ingested feeds) and in-situ nylon bag degradation characteristics (W, D and U fractions of DM, OM, CP, starch NDF) of the maize silages.

2.6 Statistical analysis

The effects of maize silage maturity, concentrate type and lactation stage on intake of nutrients, milk yield, milk composition and body condition were determined by repeated measurement analysis of variance using the PROC MIXED procedure (Littell et al., 2006) of the Statistical Analysis Systems (SAS® 2003) program. Weeks of lactation were used as a repeated effect on individual cows. Maize silage maturity, concentrate type and lactation stage were fixed effects and block was considered as a random effect. Interactions were non-significant and therefore excluded from the model:

$$Y_{ijkl} = \mu + M_i + C_j + W_k + e_{ijkl}$$

Where:
- Y_{ijkl} is the dependent variable;
- μ the general mean;
- M_i is the fixed effect of maize silage (i = MS30, MS34, MS38 and MS42);
- C_j is the fixed effect of concentrate type (j = HC and LC);
- W_k, is the fixed effect of the repeated measures of lactation weeks (k = 1-15);
- e_{ijkl} is the residual variance.

The different covariance structures of repeated matrices were evaluated according to Littell et al. (1998) and Wang and Goonewardene (2004) using the Akaike information criterion (AIC) and the Schwarz Bayesian criterion (BIC). Based on the AIC and BIC values, the unstructured covariance structure or ANTE (1) covariance structure were used in the models.

3 Results and discussion

3.1 Feed composition

Data on chemical composition and feeding values of roughage ingredients and concentrates are summarized in Table 3. The starch content of the maize silages increased (381 to 433 g/kg DM), whereas the NDF content decreased (366 to 341 g/kg DM) consistently in silages originating from the successive harvests. The content of CP, NE_L and OMD were similar across the maize silages. The variation in maturity at harvest has shown marked influence on the carbohydrate composition (starch:NDF ratio). These results are in accordance with observations in literature. The crop is harvested at an advanced ripening stage (for high starch content), but with a wide range in stage of maturation (Phipps et al., 2000; Cone et al., 2008). The increase in starch content with each subsequent harvest is related to the growth of ear and deposition of starch in the grains during maturation (Cone et al., 2008). The substantial increase in starch (grain) content decreased the NDF content in the whole crop DM. The NDF content of the stover increases as maturity advances, however, the relative NDF content of the whole crop decreases because of the simultaneous increase of the proportion of grains in whole crop DM (Bal et al., 2000). The observed starch content of maize silages were high compared to the table values of CVB (2007) and findings in other studies (Bal et al. 1997, Phipps et al. 2000). The feed stuff table of CVB indicates an average 304 g starch/kg DM for maize silage harvested in the range of 280 to 320 g DM/kg and, and 354 g starch/kg DM for maize silage harvested above 320 g DM/kg. Phipps et al. (2000) found 309 g and 354 g starch/kg DM in maize silages harvested at 330 g DM/kg and 380 g DM respectively. In the study of Bal et al. (1997), maize silage with DM contents of 301, 324, 351 and 420 contained 182, 287, 372, and 374 g starch/kg DM respectively.

Table 3 Chemical composition, and feeding value of roughage ingredients and concentrates

Parameter	Roughage mixture[1]						Concentrate[2]	
	Maize silage[3]				Grass silage	Soybean meal	LC	HC
	MS30	MS34	MS38	MS42				
Chemical composition, g/kg DM								
Dry matter (DM), g/kg	341	354	376	410	286	879	897	897
Crude protein	74	77	79	78	136	385	207	196
Crude fat	35	35	36	36	42	32	54	41
WSC [4]	4.2	4.3	4.0	4.0	58	113	90	119
Starch	381	396	415	433	-	23	94	86
NDF	366	350	345	341	487	146	386	312
ADF	212	202	198	196	289	96	213	191
ADL	19	19	19	19	17	6	41	38
pH	3.8	3.9	4.2	4.01				
NH_3-N [5], g/100 g total N	7.7	9.5	8.1	9.5	8.4	-	-	-
Feeding value [6]								
DVE [7]	50.8	51.8	51.5	50.8	48.2	208	122	123
OEB [8]	-32.3	-31.5	-29.0	-30.7	39.4	133.0	41.0	38.0
NE_L [9], MJ/kg DM	6.57	6.55	6.52	6.54	6.31	7.96	7.29	7.27
OMD [10] (%)	75.9	76.1	75.7	75.6	77.7	89.9	82.10	84.30

[1] Roughage mixture contained (DM basis): 61% maize silage, 28% grass silage, 10% soybean meal, 0.45% mineral and vitamin mixture and 0.34% salt (NaCl)
[2] Concentrates with low degradable carbohydrates (LC) or high degradable carbohydrates (HC)
[3] DM content of 300 (MS30), 340 (MS34), 380 (MS38) and 420 (MS42) g/kg fresh weight
[4] Water soluble carbohydrates
[5] Ammonia Nitrogen
[6] Calculated according to CVB (2007)
[7] Intestinal digestible protein (Tamminga et al., 1994)
[8] Degraded protein balance in the rumen (Tamminga et al., 1994)
[9] Net energy lactation calculated with VEM (feed unit lactation) system (Van Es, 1978)
[10] Organic matter digestibility determined in vitro according to Tilly and Terry (1963) as modified by van der Meer (1987)

3.2 Nutrient intake and animal performance

Intake of DM, CP and NE$_L$ did not differ (P>0.05) with advancing maturity from MS30 to MS42 (Table 4). The intake of starch increased (P<0.01), and intakes of NDF and ADF decreased (P<0.05). The intake of DM and NE$_L$ did not vary with concentrate type (Table 4). However, the intake of WSC was higher (P<0.001) and that of NDF was lower (P<0.001) on HC compared to LC concentrate due to the difference in concentrate composition. The lack of differences in DM intake is supported by earlier findings (Bal et al., 2000; Phipps et al., 2000). The results indicate that DMI is numerically but not significantly compromised at increased harvest maturity. The results suggest that the satiety value system (Zom, 2012), overestimates the adverse effects of a high DM content (>360 g DM/kg) on DMI.

Table 4 Effect of maize silages (MS) ensiled at different maturities in combination with a low (LC) or high (HC) degradable carbohydrate concentrate (Con) on nutrient intake of dairy cows

Parameter	Maize silage[1]				SEM[3]	Concentrate		SEM	Significance[2]		
	MS30	MS34	MS38	MS42		LC	HC		MS	Con	Week
Intake, kg/d											
Maize silage DM	9.7	10.0	10.0	9.5	0.38	9.9	9.7	0.35	ns	ns	***
Roughage DM	15.8	16.4	16.2	15.5	0.62	16.1	15.9	0.57	ns	ns	***
Total DM	23.2	23.5	23.7	22.8	0.68	23.4	23.1	0.64	ns	ns	***
Crude protein	3.40	3.47	3.53	3.40	0.089	3.52	3.38	0.085	#	***	***
Crude fat	0.92	0.93	0.94	0.92	0.026	0.98	0.87	0.024	ns	***	***
Starch	4.42[a]	4.71b	4.89c	4.89c	0.162	4.68	4.68	0.150	**	ns	***
WSC[4]	1.27	1.27	1.29	1.24	0.028	1.16	1.38	0.026	#	***	***
NDF	8.58[a]	8.45a	8.39a	8.06b	0.238	8.72	8.02	0.233	*	***	***
ADF	5.00[a]	4.93a	4.88a	4.70b	0.139	5.01	4.75	0.130	*	**	***
DVE[5]	1.95	1.99	1.97	1.93	0.049	1.97	1.95	0.046	ns	ns	***
NE$_L$[6], MJ/d	160	162	159	156	4.4	161	159	4.1	ns	ns	***

[1] Dry matter contents of 300 (MS30), 340 (MS34), 380 (MS38) and 420 (MS42) g/kg fresh matter
[2] ns, not significant; #, P<0.1; *, P<0.05, **, P<0.001;***, P<0.001
[3] Standard error of the mean
[4] Water soluble carbohydrates
[5] Intestinal digestible protein (Tamminga et al., 1994)
[6] Net energy lactation calculated using VEM (feed unit lactation) system 1 VEM = 6.9 kJ kJ NE$_L$(Van Es, 1978)

No difference in milk yield or milk composition was found between the maize silages, except for yield of fat, which significantly declined (P<0.05) from MS38 to MS42 (Table 5). Yield (kg/d) of milk, milk protein and milk lactose was higher (P<0.05) on the LC ration compared to the HC ration. With advance of lactation, milk yield and composition were significantly affected (P<0.001). The percentage fat and (to a smaller extent) lactose in milk was higher (P<0.05) on the HC ration compared to the LC ration. Change in body weight and BCS over the 15 weeks period did not differ with maturity of maize silages or carbohydrate composition of the concentrates. The observations regarding milk yield, body weight change and body condition in relation to harvest maturity are in agreement with previous published work (Bal et al., 2000; Phipps et al., 2000). The increase in starch:NDF ratio resulted in the numerical decrease in milk fat content from 4.25% on MS30 to 4.05% on MS42. Unexpectedly, the combination of HC concentrate with maize silages resulted in a high percentage of milk fat compared to LC concentrate (4.30% vs. 4.03%). Typically, the combination of the high fermentable carbohydrate concentrate and the low NDF roughage diets are associated with a reduction in milk fat (Nielsen et al., 2006). The high milk fat content with HC concentrate may be due to the large (2 kg/d) decrease in milk yield, as the daily fat yield did not differ between the HC and LC concentrates.

Table 5 Milk production, milk composition and changes in body condition of dairy cows fed maize silages (MS) ensiled at different stages of maturity in combination with a low (LC) and high (HC) degradable carbohydrate concentrate (Con) during week 2 to 15 of lactation

Parameter	Maize silage[1]				SEM[3]	Concentrate		SEM	Significance[2]		
	MS30	MS34	MS38	MS42		LC	HC		MS	Con	Wk
Milk yield											
Milk, kg/d	40.2	40.8	40.8	39.5	1.32	41.3a	39.3b	0.94	ns	*	***
FPCM[4], kg/d	42.9	43.4	43.8	41.6	1.45	43.2	42.6	1.02	ns	ns	***
Milk composition											
Fat, %	4.25	4.17	4.21	4.05	0.097	4.03b	4.30a	0.079	ns	***	***
Fat, kg/d	1.66ab	1.70a	1.70a	1.60b	0.067	1.67	1.67	0.047	**	ns	***
Protein, %	3.27	3.22	3.28	3.29	0.071	3.25	3.28	0.048	ns	ns	***
Protein, kg/d	1.31	1.33	1.31	1.27	0.060	1.33a	1.28b	0.058	ns	*	***
Lactose, %	4.66	4.62	4.72	4.64	0.028	4.64b	4.68a	0.024	ns	*	***
Lactose, kg/d	1.85	1.99	1.90	1.82	0.081	1.91a	1.82b	0.077	ns	**	***
Body condition											
Body weight, kg	626	640	652	650	16.3	638	646	11.5	ns	ns	ND[5]
BCS[6]	2.7	2.7	2.9	3.0	0.05	2.8	2.8	0.04	ns	ns	ND[5]

a,b Means within rows with different superscripts differ (P<0.05)
[1] Dry matter contents of 300 (MS30), 340 (MS34), 380 (MS38) and 420 (MS42) g/kg fresh matter
[2] ns, not significant; #, P<0.1; *, P<0.05, **, P<0.001;***, P<0.001
[3] Standard error of the mean
[4] Fat and protein corrected milk
[5] Not determined
[6] Body condition score on a scale of 1 to 5 according to Edmonson et al., 1989

3.3 Silage preservation

The temperature recordings of the maize silage clamps are displayed in Figure 1. The average mean temperatures were 15.4, 17.2, 18.5 and 20.3°C for silage MS30, MS34, MS38 and MS42, respectively. Although there were numerical differences in temperature between silages, the temperature was close to the ambient temperature, indicating that there was no heating. However, a sudden rise in temperature of MS42 was observed shortly before the temperature measurements were terminated (Figure 1). Measurements done in a subsequent study indicate that this sudden rise in temperature did not cause heating. The observed sudden rise in temperature was probably a matter of coincidence, possibly related to variation within the silage clamp.

Measurements at the silage blocks showed that the packed density of the maize silages in the silage bunker were 671, 611, 679 and 605 kg per m3, corresponding with 198, 208, 286 and 255 kg DM per m^3 for silage MS30, MS34, MS38 and MS42, respectively. The silage densities as observed in our study are corresponding with data from commercial and experimental farms (Van Schooten and Van Dongen, 2007), indicating that our observations could be representative for farm practice. The density of the silages was highest in MS38. Dry matter contents below or above 380 g/kg resulted in lower densities. This is also in agreement with results of Van Schooten and Van Dongen (2007).

The results show that the temperature of the silage blocks increased with time after cutting from the clamps. The temperatures were higher when measured 30 cm below the top surface compared to 40 cm above ground level (= approximately 80 cm below the top-surface), except for silage M30 which had similar temperature at the different heights (Figure 2). The differences in (the change of) silage temperature between silage MS34, MS38 and MS42 were small. The silage temperature of MS30 was numerical lower than the temperature of the other silages.
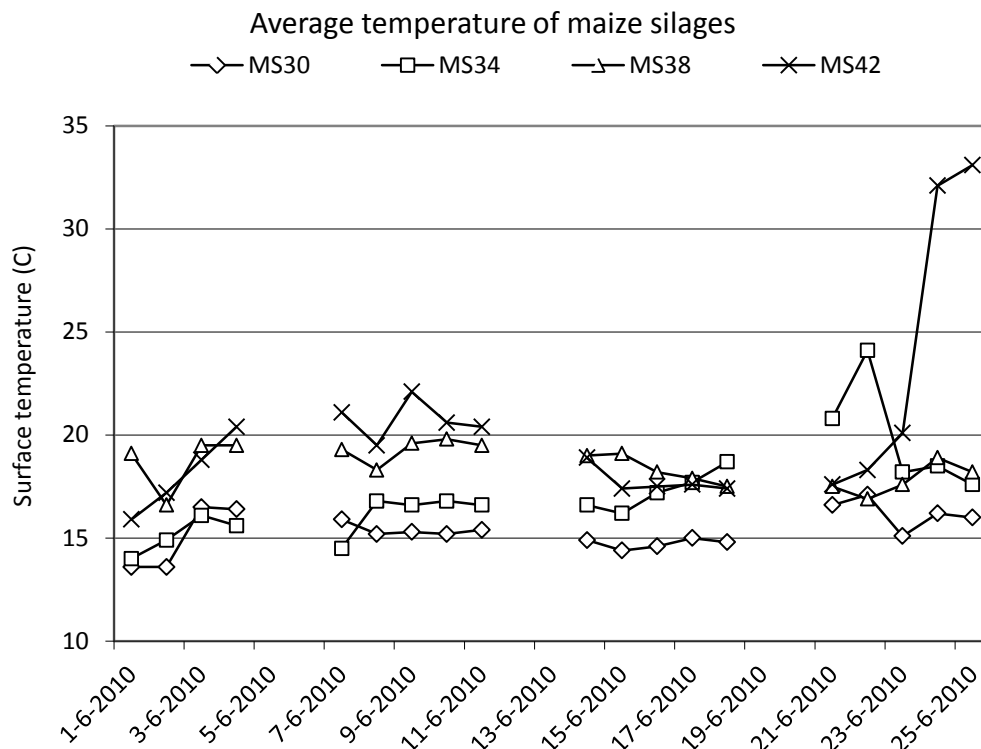
Average temperature of maize silages

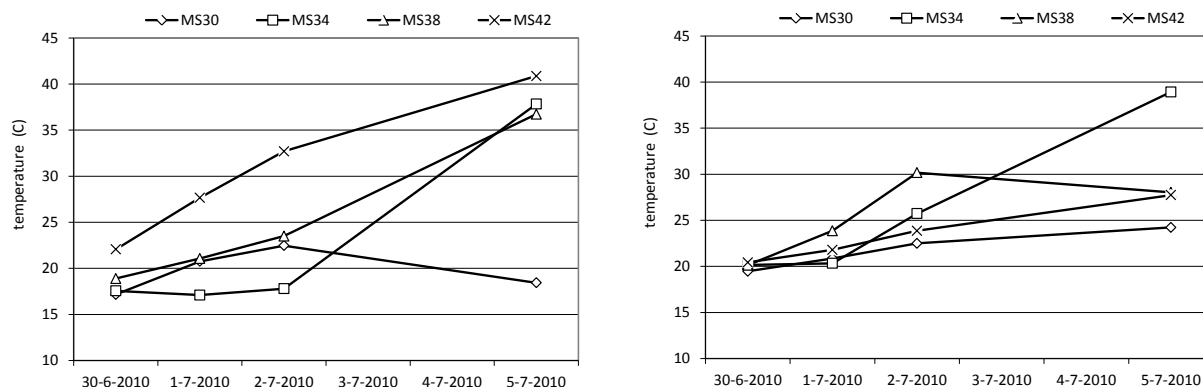**Figure 1** Average temperature of the silages, measured 30 cm under the front surface

Figure 2 Kernel temperatures of the silage blocks measured 30 cm below the upper surface (left graph) and 40 above ground level (=80 cm below upper surface; right graph)

3.4 In-situ ruminal and intestinal mobile nylon bag degradability

The results of the nylon bag degradation analysis are presented in Table 6. There were little differences in NDF digestibility between the different maize silages. The data indicate that the washable fraction of DM, OM, N and starch is reduced in MS42, compared to the other silages. The degradation rates of starch were variable, without a clear relationship with harvest maturity. Increased maturity is associated with a reduced washable fraction and an increased digestible non-washable fraction (Philippeau and & Michalet-Doreau, 1997), together with a reduced degradation rate (Flachowsky et al., 1993, Jochmann et al., 1999, Philippeau & Michalet-Doreau, 1997). Therefore, the observed increase of the washable fraction of starch in MS38 was unexpected. However, Zom (2006) did not find either a clear relationship between the size of the washable, degradable and undegradable fractions. The variability is probably related to differences in the procedures of the in-situ rumen incubation. In this study and in the study of Zom (2006), fresh and chopped samples were used according to the standard protocol, instead of dried and ground samples as used by Philippeau and

Michalet-Doreau (1997), Flachowsky et al. (1993) and Jochmann et al. (1999). Dried and ground samples have the advantage of being more homogenous and less variable than fresh chopped samples, but the disadvantage of overestimating degradation rates and the proportion of the washable (W) and the degradable (D) fraction while underestimating the outflow of starch from the rumen. However, a larger washable fraction was accompanied by a lower degradation rate. As a result, the proportion of rumen degradable starch was similar for maize silages MS30, MS34 and MS38, and slightly lower for MS42 (Table 7 and 8). Combined with the increasing starch concentration of increased harvest maturity, this resulted in an increased amount of starch digested in the rumen (or the outflow of starch from the rumen) with increased harvest maturity.

The results of the mobile nylon bag study indicated that starch from the maize silages entering the duodenum was almost entirely digested in the ileum. The ileal residues of starch were 1.47, 1.58, 1.77 and 1.57%. The calculated amount of starch entering the large intestine was very low with 26, 29 34 and 33 g/day for maize silage MS30, MS34, MS38 and MS42, respectively (see also Table 7 and Table 8).

Table 6 In-situ nylon bag degradation 1) of dry matter (DM), organic matter (OM), nitrogen, starch and NDF in maize silages harvested at different crop maturities 300 g DM/kg fresh weight (MS30); 340 g DM/kg fresh weight (MS34); 380 g DM/kg fresh weight (MS38); and 420 g DM/kg fresh weight (MS42

Maize silage		DM	OM	N	Starch	NDF	NDF lag time
MS30	W (%)	26.7	25.2	57.6	36.7	0.0	0.0
	U (%)	9.6	9.2	14.6	0.2	19.0	19.8
	D (%)	63.7	65.7	27.9	63.1	81.0	80.2
	kd (%/h)	3.6	3.7	2.8	7.2	1.9	2.3
	lag-time (h)	0.0	0.0	0.0	0.0	0.0	6.2
MS34	W (%)	24.5	22.4	58.8	27.9	0.0	0.0
	U (%)	11.0	9.3	15.9	0.2	19.1	19.9
	D (%)	64.5	68.3	25.3	71.9	80.9	80.1
	kd (%/h)	4.0	4.1	2.5	9.8	2.0	2.2
	lag-time (h)	0.0	0.0	0.0	0.0	0.0	4.0
MS38	W (%)	27.2	25.8	58.1	37.5	0.0	0.0
	U (%)	9.8	9.4	14.7	0.3	21.0	21.9
	D (%)	63.0	64.8	27.2	62.2	79.0	78.1
	kd (%/h)	3.7	3.8	2.5	6.0	2.0	2.3
	lag-time (h)	0.0	0.0	0.0	0.0	0.0	6.9
MS42	W (%)	18.6	16.6	44.0	7.7	0.0	0.0
	U (%)	11.0	10.4	18.5	0.1	23.6	24.3
	D (%)	70.4	72.9	37.5	92.2	76.4	75.7
	kd (%/h)	3.7	3.8	2.8	6.9	1.7	2.0
	lag-time (h)	0.0	0.0	0.0	0.0	0.0	5.5

[1]   Data fitted on the model of Ørskov & McDonald (1979), $Y(t)=W+D \times (1-e^{kd \times t})$
[2]   W = washable fraction, washout from the nylon bags
[3]   U = undegradable fraction, undegraded after 336 h rumen incubation
[4]   D = (100-W-U) potential rumen degradable fraction
[5]   kd (%/h) = degradation rate
[6]   Lag-time = time elapsed between incubation and start of degradation

3.5 Model simulation

The results of the model simulation of starch digestion and enteric methane emission as affected by harvest maturity and concentrate type are presented in Tables 7 and 8. The outflow of starch from the rumen to the intestine tends to increase with increasing harvest maturity. Although the absolute amount of starch digested in the rumen was simulated to increase, the proportion of starch digested in the rumen decreased with increasing harvest maturity, indicating a shift in starch digestion from the rumen to the intestine.

Table 7 demonstrates the effect of the increased maturity at harvest and the shift of starch digestion from the rumen to the intestine on methane emission, assuming an identical DMI of 23.0 kg/d, hence

excluding effects of DMI. The results in Table 7 indicate that the predicted methane conversion factor and the enteric methane emission (g CH_4/d) both slightly decrease (2%) with advancing maturity at harvest. Table 8 shows the effect of increased maturity at harvest using the observed DMI. From Tables 7 and 8 it can be concluded that the impact of DMI and the effects of differences in starch digestibility have a similar impact on the enteric emissions of methane. For all treatments, the CH_4 conversion factor (percentage of gross energy converted to methane) was lower than the fixed value of 0.065 as used according to the TIER 2 approach of the IPCC. The predicted enteric methane production per unit FPCM remained similar for all treatments.

The effect of harvest maturity of maize on the predicted enteric emission of methane was lower than suggested by Tamminga et al. (2007). This is probably due to the relatively high starch content at each stage of harvest maturity. As pointed out earlier, the starch contents in our study were much higher than reported in literature (Bal et al., 1997, Phipps et al. 200, CVB, 2007), whereas the effect of an increased harvest maturity on the starch content of the silage was small. Increasing harvest maturity from 330 to 420 g DM resulted in an increase in starch content of only 14%, which is much lower than the effects reported by Phipps (2000) and Bal et al. (1997). The latter study showed an increase in starch content from 182 to 374 g starch/kg DM when harvest maturity was increased from 301 to 420 d DM/kg, which is an increase of 105%. The small effect of harvest maturity on starch content may be related to the favourable growing conditions. Efforts of maize breeders may also have contributed to higher starch contents in the present study compared to contents reported in literature. Starch content is an important criterion of acceptance of new maize varieties on the recommended maize variety list. Therefore, breeders put much effort into increasing the starch content of maize silage.

Table 7 Results of the model simulation of enteric CH_4 production as affected by maize silage and concentrate type, assuming an identical dry matter intake of 23.0 kg/d

	Harvest maturity[1]				Concentrate[2]	
	MS30	MS34	MS38	MS42	LC	HC
Feed intake (kg DM /d)	23.0	23.0	23.0	23.0	23.0	23.0
Gross Energy intake (MJ/d)	420	419	420	421	422	418
Methane emission						
Methane Conversion Factor (% of GE)	5.64	5.67	5.56	5.53	5.54	5.66
Methane production (g/d)	425	425	423	418	5.56	5.65
Methane (g/kg DM)	18.3	18.5	18.4	18.2	18.3	18.5
Starch digestibility[3]						
Rumen digested starch (%)	66.3	67.5	65.9	61.7	64.7	65.4

[1] Dry matter content at harvest MS30 = 300 g DM/kg fresh weight, MS34 = 340 g DM/kg fresh weight, MS38 = 380 g DM/kg fresh weight, MS42 = 420 g DM/kg fresh weight

[2] Concentrate LC = 90 g/kg DM water soluble carbohydrates,386 g NDF; HC = 119 g water soluble carbohydrates 312 g NDF

[3] Results derived from in situ nylon bag degradation and mobile nylon bag degradation

Table 8   Results of the model simulation of enteric CH_4 production as affected by maize silage and concentrate type, with observed dry matter intake, averaged per experimental group

	Harvest maturity[1]				Concentrate[2]	
	MS30	MS34	MS38	MS42	LC	HC
Feed intake (kg DM /d)	23.2	23.5	23.7	22.8	23.4	23.1
FPCM yield (kg/d)	42.9	43.4	43.8	41.6	43.2	42.6
Gross Energy intake (MJ/d)	420	430	435	419	429	423
Methane emission						
Methane Conversion Factor (% of GE)	5.64	5.67	5.56	5.53	5.54	5.66
Methane production (g/d)	426	434	435	416	427	428
Methane (g/kg DM)	18.3	18.5	18.4	18.2	18.3	18.5
Methane (g/kg FPCM)	9.9	10.0	10.0	10.0	9.9	10.0
Starch digestibility[3]						
Rumen digested starch (%)	66.3	67.0	65.2	61.9	64.7	65.4
Ileal digested starch (%)[4]	98.5	98.4	98.3	98.4	98.4	98.4
Starch entering large intestine (g/d)	26	29	34	33	30	31

[1] Dry matter content at harvest MS30 = 300 g DM/kg fresh weight, MS34 = 340 g DM/kg fresh weight, MS38 = 380 g DM/kg fresh weight, MS42 = 420 g DM/kg fresh weight
[2] Concentrate LC = 90 g/kg DM water soluble carbohydrates,386 g NDF; HC = 119 g water soluble carbohydrates 312 g NDF
[3] Results derived from in situ nylon bag degradation and mobile nylon bag degradation
[4] Percentage of by-pass starch digested in the small intestine

4 Conclusions

The maturity of maize silages at harvest (300 to 420 g/kg fresh weight) did not affect the dry matter intake and the yields of milk, FPCM, milk protein and milk lactose, or body condition score. The milk fat yield was depressed at a harvest maturity of 420 g DM/kg. There were no interacting effects of concentrate composition vs. maize silages on DMI and milk performance, indicating that the results from this study are valid for a wide range of practical situations with different types of concentrates.

The results of this study indicate that increasing the harvest maturity of silage maize above 350 g DM/kg increases starch intake as well as the proportion of rumen by-pass starch, resulting in an increased flow of starch to the intestine. In-situ mobile nylon bag data demonstrated that the total amount of rumen by-pass starch was entirely digested in the small intestine, showing that increasing the harvest maturity of silage maize did not increase hind gut fermentation of faecal starch loss.

Nevertheless, harvest maturity of maize silage had a reducing effect on the calculated enteric methane emission in early lactation. Compared to values reported in literature, the starch contents of all maize silages used in the present study were much higher, and the differences between the different stages of harvest maturity was much smaller than expected. As a result, the effects of increased harvest maturity on the starch content and the calculated methane emission remained smaller than expected.

Increasing the harvest maturity of silage maize had no effects on the stability of the silages. With appropriate silage management there is no increased heating or moulding of the maize silages with increasing dry matter content.

This study confirms that increasing harvest maturity of silage maize can be managed without adverse effects on animal performance, silage digestibility or silage preservation. With appropriate silage management such as good compaction, use of a sand load on top of the clamp and a sufficient rate of feed removal from the clamp (0.15 m/d), heating and moulding can be avoided. The current recommendation to harvest silage maize within the range of 300 to 360 g DM/kg can be stretched to a wider range of 300 to 420 g DM/kg. As increasing harvest maturity may reduce methane emission, especially in maize cultivars with lower starch content at early harvest, delayed harvest is a feasible "no cost" and "no regret" measurement to reduce the enteric methane emission from dairy farming.

Literature

Bannink, A., M.W. van Schijndel and J. Dijkstra. 2011 A model of enteric fermentation in dairy cows to estimate methane emission for the Dutch National Inventory Report using the IPCC Tier 3 Approach. *Anim. Feed Sci. Techn.* 166-167, 603-618.

Bal M.A., J.G. Coors and R.D. Shaver. 1997. Impact of the Maturity of Corn for Use as Silage in the Diets of Dairy Cows on Intake, Digestion, and Milk Production. *J. Dairy Sci.* 80, 2479-2503.

Bal M.A., R.D. Shaver, A.G. Jirovec, K.J. Shinners and J.G. Coors. 2000. Crop processing and chop length of corn silage: Effects on intake, digestion, and milk production by dairy cows. *J.Dairy Sci.* 83, 1264-1273.

Barrière Y., D. Alber, O. Dolstra, C. Lapierre, M. Motto, A. Ordas, J. Van Waes, L. Vlasminkel, C. Welcker and J.P. Monod. 2006. Past and prospects of forage maize breeding in Europe. II. History, germplasm evolution and correlative agronomic changes. *Maydica* 51, 435-49.

Cone J.W., A.H. Van Gelder, H.A. Van Schooten and J.A.M. Groten. 2008. Effects of forage maize type and maturity stage on in vitro rumen fermentation characteristics. *Neth.J.Agric.Sci.* 55, 139-154.

CVB, 2003. Protocol voor in situ pensincubatie - Bepaling van afbraaksnelheid en uitwasbare fracties van eiwit,zetmeel, celwanden en organische restfractie. Centraal Veevoederbureau, Lelystad, Nederland, pp. 14.

CVB, 2007. Table Ruminants, chemical composition and nutritional values of feedstuffs and feeding standards, Series no 32. Central Bureau for livestock feeding (CVB), the Hague, the Netherlands.

Edmonson A.J., I.J. Lean, L.D. Weaver, T. Farver and G. Webster. 1989. A body condition scoring chart for Holstein dairy cows. *J.DairySci.72*, 68-78.

Folch J., M. Lee and G.H.S. Stanley. 1957. A simple method for the isolation and purification of total lipids from animal tissues. *J.Bioch.Chem.226*, 497-509.

ISO, 2005. Animal feeding stuffs - Determination of nitrogen content and calculation of crude protein content - Kjeldahl method. International Organsation for Standardisation, Geneve, Switzerland.

Khan N.A., J.W. Cone and W.H. Hendriks. 2009. Stability of fatty acids in grass and maize silages after exposure to air during the feed out period. *Anim.Feed Sci.Technol.154*, 183-192.

Littell R.C., P.R. Henry and C.B. Ammerman. 1998. Statistical Analysis of Repeated Measures Data Using SAS Procedures. *J.Anim.Sci.* 76, 1216-1231.

Littell R.C., G.A. Milliken, W.W. Stroup, R.D. Wolfinger and O. Schabenberger. 2006. SAS System for Mixed Models (2nd edn). SAS Institute, Cary, NC. USA.

Norberg, E., H. Volden and O.M. Harstad. 2007. Technical Note: Assessment of Recovery Site of Mobile Nylon Bags for Measuring Ileal Digestibility of Starch in Dairy Cows. *J.DairySci.90*: 418-421.

O'Mara F.P., J.J. Fitzgerald, J.J. Murphy and M. Rath. 1998. The effect on milk production of replacing grass silage with maize silage in the diet of dairy cows. *Livest.Prod.Sci.* 55, 79-87.

Ørskov, E.R. and I. McDonald. 1979. Estimation of Protein Degradability in the Rumen from Incubation Measurements Weighted According to Rate of Passage. *J.Agric. Sci.* 92, 499-503.

Phipps R.H., J.D. Sutton, D.E. Beever and A.K. Jones. 2000. The effect of crop maturity on the nutritional value of maize silage for lactating dairy cows 3. Food intake and milk production. *Anim.Sci.71*, 401-409.

Phipps R.H., J.D. Sutton and B.A. Jones. 1995. Forage mixtures for dairy cows: The effect on dry-matter intake and milk production of incorporating either fermented or urea-treated whole-crop wheat, brewers' grains, fodder beet or maize silage into diets based on grass silage. *Anim.Sci.* 61, 491-496.

Scheiner D. 1976. Determination of ammonia and Kjeldahl nitrogen by indophenol method. *Water Res.10*, 31-36.

Schroeder J. 1998. Towards improved nitrogen management in silage maize production on sandy soils. PhD Thesis Wageningen University, The Netherlands.

Sibma L, Growth and Development of Maize (Zea mays L.) under Dutch Conditions. Pudoc, Wageningen (1987). (in Dutch).

Struik P.C. 1983. Effect of temperature on development, dry-matter production, dry- matter distribution and quality of forage maize (Zea mays L.): an analysis. PhD Thesis Wageningen University, The Netherlands.

Tamminga S., A. Bannink and R. L.G. Zom. 2007. Feeding strategies to reduce methane loss in cattle Wageningen UR Animal Sciences Group, Lelystad, Rapport 34 pp. 46

Tamminga S., W.M. Van Straalen, A.P.J. Subnel, R.G.M. Meijer, A. Steg, C.J.G. Wever and M.C. Blok. 1994. The Dutch protein evaluation system: the DVE/OEB-system. *Livest.Prod.Sci.* 40, 139-155.

Tilley J.M.A. and R.A. Terry. 1963. A two-stage technique for the in vitro digestion of forage crops. *J. Br. Grassl. Soc.* 18, 104-111.

Van Dijk, W., J. Groten, W. van den Berg, J. Kassies and J. Weijers. 2006. Efficiënt gebruik van snijmaïs. Deel 2: invloed van rastype en oogststadium op opbrengst en kwaliteit. Wageningen UR Animal Sciences Group, Lelystad, PraktijkRapport Rundvee 85 pp. 51.

Van Es A.J.H. 1978. Feed evaluation for ruminants. I. The systems in use from May 1977-onwards in The Netherlands. *Livest. Prod. Sci.* 5, 331-345.

Van Schooten, H.A. 2006. Efficiënt gebruik van snijmaïs. Deel 3: invloed van rastype en oogststadium op conservering Wageningen UR Animal Sciences Group, Lelystad, PraktijkRapport Rundvee 86. pp 23.

Van Schooten H.A. and C.A. van Dongen. 2007 Dichtheidsbepaling maïs- en graskuilen met boormonsters. Wageningen-UR Animal Sciences Group, Lelystad, Nederland Rapport 64 pp 23.

Van Soest P.J. 1973. Collaborative study of acid detergent fibre and lignin. *J. Assoc.Off. Anal.Chem.* 56, 781-784.

Van Soest P.J., J.B. Robertson and B.A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber and non-starch polysaccharides in relation to animal nutrition. *J.Dairy Sci.* 73, 2583-3593.

Van Vuuren A.M., C.J. van der Koelen, H. Valk and H. de Visser. 1993. Effects of partial replacement of ryegrass by low protein feeds on rumen fermentation and nitrogen loss by dairy cows. *J. DairySci.* 76, 2982-2993.

Volden, H. and O.M. Harstad. 1995. Effect of rumen incubation on the true indigestibility of feed proteins in the digestive tract determined by the nylo bag techniques. *Acta Agric.Scan.Section A Anim.Sci.* 45, 106-115.

Wang Z. and L.A. Goonewardene. 2004. The use of MIXED models in the analysis of animal experiments with repeated measures data. *Can.J. Anim.Sci.* 84, 1-11.

Zom, R.L.G. 2006 Efficiënt gebruik van snijmaïs Deel 5: invloed afrijpingstype en oogststadium op opname en melkproductie. Wageningen-UR Animal Sciences Group PraktijkRapport 88 pp. 43