REFEREED ARTICLE

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Maize cultivars for anaerobic digestion and animal nutrition in Europe

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ABSTRACT

Increased use of whole-plant maize for anaerobic digestion (AD) in Europe raises the question: Are maize cultivars developed for use in animal nutrition equally appropriate as feedstock for AD or should different phenotypes be selected? The main objective in growing whole-plant maize as feedstock for AD is maximum output of methane per hectare. There is less need for rapidly digested plant components such as starch in AD feedstock than in a ruminant diet because the typical digestion period is several weeks for AD compared with less than two days for the rumen. The ideal phenotype of maize for AD is a very high yielding plant with a low lodging score. Metabolisable energy (ME) intake from forage is a limiting factor to output of animal product per head, thus, in addition to high dry matter yield per hectare, a high concentration of ME in the maize plant is desirable. Major factors contributing to high ME in whole-crop maize are starch and digestible plant cell wall. The ideal phenotype of maize for animal nutrition is therefore a plant with a high proportion of ear, a low concentration of lignin, high cell wall digestibility and low lodging score.

KEYWORDS: Forage maize; phenotype; anaerobic digestion; biogas; animal feed; methane

1. Introduction

Ensiled whole-plant maize (*Zea mays*) is widely used throughout the world as animal feed. However, increasing quantities of the crop have been grown in Europe in recent years specifically as feedstock for anaerobic digestion (AD) for the production of methane biogas. In Germany, for example, of the total area of 3.6 million hectares of maize planted in 2014, 0.50 was for silage for animal feed, 0.33 was destined for biogas 0.17 was grown for grain (H. Messner, personal communication). In UK some 15,500 hectares of forage maize was grown in 2013 as feedstock for biogas, 0.10 of the total maize area (National Institute of Agricultural Botany, 2013). The UK Descriptive List of forage maize cultivars nominated as having potential suitability for AD use includes separate lists for favourable and less favourable sites, with details for each cultivar of concentrations of dry matter (DM) and metabolisable energy (ME) together with yield of DM and ME, early vigour and standing power at harvest or root lodging (NIAB 2015). These characteristics are similar to those assessed for maize cultivars in the UK Descriptive List of forage maize cultivars for animal feed (NIAB, 2015).

Feedstock and feed inputs comprise the major variable costs of biogas and livestock production. For example, cost of feedstock was estimated to comprise 0.49 to 0.83 of total variable costs of farm-scale AD (Redman, 2010) and the cost of animal feed comprised 0.76 of the total variable costs of milk production (DairyCo, 2014). Since forage has a lower unit cost than concentrate feed (DairyCo, 2012), optimum output of livestock product from forage is a key performance objective. For example, a target for milk production is for forage energy intake to comprise 0.50 of total annual energy intake (Wilkinson, 2013).

Two major operational objectives in AD are maximisation of specific methane (CH_4) yield (litres of CH_4 per kg volatile solids, VS) and maximum methane yield per hectare of land (Amon *et al*., 2007b). In contrast, a major objective of most livestock production is the optimisation of daily output of milk or live weight gain per animal within the constraints of input costs, especially when factors other than land such as labour or animal accommodation are the primary limiting resources.

The development of methane biogas production on a farm-scale using ensiled whole-plant maize as the sole or primary feedstock raises the question: Are maize cultivars developed for ensiling as animal feed equally well-suited for use as feedstock for AD and if not what phenotype of maize should be selected specifically for AD? It could be argued that maximising energy yield per hectare is an important objective in the production of both biogas and animal feed. This may be correct, provided specific methane yield per kg volatile solids and the concentration of ME per kg DM are not compromised by choosing a variety of maize of high DM yield but low methane yield or ME per kg VS or DM, or harvesting at a late stage of crop maturity so

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that daily output of methane or voluntary intake of ME and animal output are reduced by more than the increase in yield of DM.

In this review factors affecting methane production are discussed, the main features of fermentation in the anaerobic digester and rumen are compared, and phenotypic traits of whole-plant maize for AD are compared with those for animal nutrition.

2. Methane production

Maximum methane production is the main objective in operating a digester. However, methane is a greenhouse gas (GHG) and emissions of methane from enteric fermentations comprise 0.39 of global livestock GHG emissions (Opio, 2013). Minimising methane production is therefore an important environmental objective in ruminant animal nutrition.

Methane is the final product of a multi-stage process. The methanogenic organisms responsible for the production of methane, the *Archaea*, do not ferment carbohydrates, proteins or lipids, but gain energy by reducing the end products of the fermentation process such as carbon dioxide, acetic acid, formic acid and methanol with methane being produced as a by-product of the reduction process (Moss, 1993). The A*rchaea* are strict anaerobes and both digester and rumen are ideal environments for their development with excess hydrogen from microbial digestion of feed producing highly reduced conditions (Eh -350 mV). Hydrogen is potentially poisonous to the microbial population and must therefore be removed. Several hydrogen sinks exist, of which methane is by far the most important. Other hydrogen sinks include the production of ammonia from the degradation of amino acids and the saturation of unsaturated fatty acids (Moss, 1993).

Typically 0.60 of total DM in ruminant diets is carbohydrate, 0.15 to 0.20 crude protein, 0.10 ash and 0.10 lipid (McDonald *et al.*, 1995). The digestion of carbohydrate (mainly cellulose, hemicellulose, starch and fructans) by the microbial population results in the production of simple sugars, mainly hexoses, which are rapidly fermented to steam-volatile fatty acids (VFA) such as acetic, propionic, butyric and valeric. At pH 6 and above VFA are present as their dissociated salts - acetate, propionate and butyrate (Penner, 2014). Protein and other nitrogenous compounds such as amides and amines are reduced to ammonia, some of which is incorporated into microbial protein (McDonald *et al.*, 1995). A key intermediate in the digestion of carbohydrate is pyruvate, which is fermented to VFA and formate. The formate is converted to carbon dioxide and hydrogen, probably by enzymes produced by methanogens.

Importantly, excess hydrogen is produced with the production of acetate and butyrate but not with the production of propionate and valerate (Moss, 1993). Feeds containing lower levels of fibre and higher proportions of starch tend to result in higher proportions of propionic and lower proportions of acetic acid than higher fibre feeds and feeds (Table 1). The proportions of different VFA vary with the relative proportions of different bacterial species - those producing acetate predominating with feeds higher in fibre and at higher fermentation pH (above pH 6.0). Thus the pattern of VFA production affects the amount of excess hydrogen and hence the amount of methane produced per mole of hexose sugar fermented (Table 1).

Table 1: Production of methane from hexose sugar fermentation in the rumen

Molar ratio of acetate: propionate: butyrate in rumen fluid	Moles of methane produced per mole of hexose fermented in rumen
70:20:10 (forage diet)	0.64
55:30:15 (concentrate diet)	0.48

Source: Moss, 1993.

Methane has a gross energy value of 55 MJ/kg DM, compared to 17.5 MJ/kg DM for cellulose and 17.7 MJ/kg DM for starch - the two major fermentable substrates in maize (McDonald *et al.*, 1995). Methane energy loss in ruminants generally accounts for about 0.05 of gross energy intake but can vary widely from 0.02 to 0.12 of gross energy intake (Holter and Young, 1992; Johnson and Johnson, 1995) - more for fibrous feeds and less for concentrate and previously fermented feeds such as brewers' grains (McDonald *et al.*, 1995). Research is currently underway to produce a wider range of methane emission factors for livestock because it is recognised that current values do not represent the full range of diets, classes of animal and systems of production currently in use on farms.

Two major factors affect the total amount of methane produced per digester or animal – the amount of feedstock or feed DM consumed daily and its digestibility (Tamminga *et al.*, 2007) with the most important factor for the animal being daily DM intake (Mills *et al.*, 2008). Early research demonstrated that the digestible energy concentration of the diet (reflecting fibre concentration and fibre digestibility) had a major influence on methane energy produced per unit of gross energy eaten (Blaxter and Clapperton, 1965). Energy balance studies with dairy cows given a wide range of diets showed that daily methane production per animal was positively related to DM intake and diet NDF concentration, and negatively related to diet concentrate proportion (Yates *et al.*, 2000). Studies with dairy cows have also shown that substitution of grass silage by maize silage reduces methane emissions (Tamminga *et al.*, 2007: Garnsworthy *et al.*, 2012), although this mitigation of methane emissions may be offset by soil carbon loss following the ploughing of grassland for maize cultivation (Vellinga and Hoving, 2011).

In AD, type of feedstock can have a major impact on specific methane yield. Typical specific methane yields for a range of feedstock are shown in Table 2. Maize silage is intermediate between manure and food waste. Crude fat (total oil) concentration in maize is related positively to specific methane yield (Rath *et al.*, 2013) and, in contrast to the rumen, feedstocks with higher concentrations of oil such as rapeseed meal and waste cooking oil yield more methane per kg volatile solids than feedstock with lower oil concentration such as maize silage (Table 2). Addition of long-chain fatty acids to the diet depresses methane production in the ruminant (Blaxter and Czerkawski, 1966), often with associated decreases in DM intake, NDF digestibility and milk production (Tamminga *et al.*, 2007).

Table 2: Specific methane yields from a range of feedstock

Feedstock	Specific methane yield (litres CH_4/kg volatile solids)
Cow manure	190
Rye	300
Maize silage	320
Wheat grain	370
Waste vegetables	380
Food waste	400
Rapeseed meal	410
Waste cooking oil	540

Source: Al Seadi *et al*, 2008.

The extent to which methane production is decreased in the animal depends on fatty acid chain length and degree of unsaturation (Giger-Reverdin *et al*., 2003), with longer chain and unsaturated fatty acids possibly having a toxic effect on gram-positive bacteria in a similar way to the action of the gram-positive antibiotic monensin, which also reduces methane and acetate but not propionate production in the rumen (Russell and Strobel, 1987). A possible explanation for the positive relationship between feedstock oil concentration and specific methane yield in AD is that the microbial population adapts to higher fatty acid feedstock during the relatively long residence time in the digester.

3. Anaerobic digester compared to rumen

To answer the question of phenotypic suitability of whole plant maize for AD or rumen digestion, it is essential to know to what extent the anaerobic digester and rumen are similar in terms of optimal operational parameters and in what respects they differ. Of fundamental importance in both AD fermentation vessel and rumen is optimisation of both physiological and biochemical conditions for microbial digestion of crop components. Typical optimal operating parameters for AD and rumen are shown in Table 3. Common features include concentration of total volatile solids in feedstock or dry matter (DM) in animal diet, optimal fermentation temperature, pH and concentration of ammonia-N.

The most important difference between digester and rumen is in residence time – on average several weeks for AD but less than 2 days for rumen, with potential consequences for optimal speed of digestion, which may be different for AD compared to animal diet. Speed of degradation of plant substrates by microbial enzymes, with production of volatile fatty acids (VFA), carbon dioxide and methane as principal fermentation end-products, determines rate of fermentation in both AD digester and rumen. Maintenance of pH above 6 is essential for maintaining fibre digestion in the rumen (Ørskov, 1998; Offer *et al*., 2004) and also for growth of methanogenic microorganisms in the digester (Weiland, 2010). Rapidly digested substrates such as starch and water-soluble carbohydrates (sugars) can result in the production of VFA at a rate that exceeds the buffering effects of salts or saliva with the result that the pH of the digester or rumen can fall. Lower rumen pH due to rapid production of VFA can predispose the animal to sub-acute acidosis (Kleen *et al*., 2003). In this situation the microbial population changes and the mix of VFA shifts from acetate towards propionate. In situations of excess ruminal acidity (below pH 5.5) the microbial population can change further with the production of lactic acid (Chamberlain and Wilkinson, 1996) with continued reduction in pH because lactate-producing bacteria are more tolerant of low pH conditions than acetogenic bacteria (McDonald *et al*., 1991).

Adequate buffering of fermentation acids is therefore vital in both digester and rumen. Offer *et al*. (2004) ascribed rumen stability values to different feeds according to concentration of neutral detergent fibre (NDF) and potential acid load (PAL), determined *in vitro* by incubating a feed for 24 hours with rumen liquor and measuring the amount of alkali required to raise the pH of the incubation mixture back to pH 7.25 (i.e. the total free acid produced by the fermentation). PAL is now estimated in grass silages routinely by near infra-red reflectance spectroscopy (NIRS) to identify silages that may increase the risk of sub-acute ruminal acidosis in the animal (Walker, 2014). It is assumed that feeds like hay with a relatively low PAL (800 meq/kg DM) effectively have a neutral effect on rumen pH in terms of their buffering capacity and fermentation acid production because the rate of acid production from their fermentation can be balanced by plant buffering constituents, salivary bicarbonate and rumen ammonia. Feeds with higher PAL such as maize silage (1000 meq/kg DM) or wheat grain (1250 meq/kg DM) tend to lower rumen pH and need more salivary bicarbonate, produced

Table 3: Typical optimal operating parameters for anaerobic digestion and rumen digestion

	Digester	Rumen
Feedstock or feed		
Total volatile solids or DM (g/kg fresh weight)	100 to 300	400 to 500
Carbon: Nitrogen	25:1	15:1
Fermentation		
Temperature (°C)	25 to 40	38 to 40
pH*	7 to 8	6 to 7
Ammonia-N (mg/litre)	50 to 70	50 to 80
Average residence time (days)	21 to 65	1 to 2
Biogas		
Methane (% of total gas)	50 to 60	30 to 40
Methane energy (% of total energy intake)	55 to 80	3 to 15

*Optimal conditions

Source: Satter and Slyter, 1974; McDonald *et al*., 1995; Chamberlain and Wilkinson, 1996; Holter and Young, 1992; Ørskov, 1998; Amon *et al*., 2007b; Al Seadi *et al*., 2008 and Weiland, 2010.

during rumination and stimulated by fibrous feeds of relatively high NDF concentration (Schultze *et al.*, 2014), to balance this effect (Offer *et al.*, 2004). In the absence of such information for different feedstocks destined for use in AD, evaluation of their potential effects on the pH of digestate in terms of PAL would be a valuable aid to feedstock formulation. However, the variation in PAL and NDF between different varieties of forage maize is likely to be relatively small compared to that between different crop species and by-products used as feedstock sources for AD.

Ammonia nitrogen (N) can accumulate in digestate and rumen when the supply of feed protein or non-protein nitrogen (e.g. urea) exceeds its assimilation into protein by the microbial population. Elevated concentrations of ammonia N (>80 mg NH_3-N/litre) can be toxic to methanogens (Al Seadi *et al.*, 2008) and give rise to raised concentrations of NH_3 in biogas (Strick *et al.*, 2006). Higher concentrations of ammonia in the rumen can lead to elevated concentrations of ammonia in blood, with increased risk of reduced livestock fertility (McEvoy *et al.*, 1997). The optimal concentration of NH_3-N in rumen fluid is 50 to 80 mg/litre (Satter and Slyter, 1974), similar to that for AD (Table 1). Forage maize has a relatively low concentration of crude protein (N x 6.25) compared to other forage crops and by-products (Thomas, 2004) and would normally be balanced by additional supplementary protein or NPN to meet requirements for degradable N (Chamberlain and Wilkinson, 1996). The risk of excess ammonia in digestate and rumen fluid is low provided supplementary N is included at the correct level, mixed uniformly with other ingredients and there are no other factors (e.g. toxins or deficiencies in essential minerals) that might reduce microbial growth and reduce the rate of synthesis of ammonia into microbial protein.

Diet formulation for the dairy cow involves balancing the composition of one ingredient with that of others so that the total diet meets the requirement of the animal for nutrients within constraints, of which the most significant is daily DM intake. Thus the relatively low protein concentration of maize silage is balanced with feeds of relatively high protein concentration such as lucerne, soyabean meal or urea. Similarly, adding complementary components to the digester can mitigate variation in individual feedstock composition. Phenotypic variance in whole-plant maize feedstock may therefore be of lesser importance than crop yield *per se* in determining choice of cultivar provided alternative sources of feedstock are available at competitive cost.

4. Desirable traits of whole-plant maize

Compared to other crops, forage maize has three important characteristics that contribute to making well-preserved silage - relatively high concentrations of DM and water-soluble carbohydrates and a relatively low buffering capacity or resistance to acidification. Thus the risk of secondary (clostridial) fermentation in maize silage is low, even at relatively low DM concentration (Weissbach *et al.*, 1974; Wilkinson, 2005). However, excessive loss of water-soluble carbohydrates, high silage acidity and elevated concentrations of soluble nitrogen are features of whole-plant maize ensiled at low concentrations of DM (Wilkinson and Phipps, 1979; Wilkinson *et al.*, 1998). It is therefore advisable to harvest the crop at DM concentrations above 275g/kg fresh weight to minimise fermentation losses.

Barrière *et al.* (1997) reviewed the phenotypic attributes of forage maize for silage and stressed the importance of a well-developed rooting system to aid resistance to lodging and drought, and also to increase efficiency of nitrogen utilisation by the crop. They suggested a target grain concentration at harvest of 0.46, corresponding to 0.30 starch, as optimal in maize silage for dairy and beef cattle. The target stage of maturity at harvest for optimal utilisation by the dairy cow is 300 to 350g DM/kg fresh weight (Browne *et al.*, 1995; Wilkinson *et al.*, 1998). At this stage of plant maturity starch comprises about 0.67 of the grain endosperm (Bal *et al.*, 1997).

Optimal maize plant maturity for AD is probably similar to that for the animal, though the decrease in plant cell wall (NDF) digestibility with advancing plant maturity may be relatively less important for AD than for the animal in view of the longer residence time in the digester than in the cow (Table 1). Nevertheless, rate of fibre digestion and residence time in the digester determine rate of methane production. Enhanced digestibility of maize silage allows average residence time in the digester to be reduced or, for new digesters, the same amount of methane may be produced from a digester with a smaller volume. Weissbach (2009) found that gas yield from a range of silages was related to digestible (i.e. fermentable) organic matter (FOM), which in turn could be predicted from concentrations of ash and acid detergent fibre. Average potential biogas yield from silages was 800 litres/kg FOM and methane yield was 420 litres/kg FOM. Frei (2013) reviewed the different roles of lignin, a complex carbohydrate polymer cross-linked to cell wall hemicelluloses that confers structural strength to the plant, in plant stress, animal nutrition and bio-energy production. He concluded that low concentrations of lignin are desirable for both animal feeding and biogas production. In the ruminant, lignin concentration and NDF digestibility are inversely related (Van Soest, 1994). Oba and Allen (1999a) found that a one unit increase in forage NDF digestibility *in vitro* was reflected in 0.17 kg increase in DM intake and 0.25 kg increase in fat-corrected milk yield in dairy cows. The lower lignin brown midrib (*bm3*) mutant (Cherney *et al.*, 1991) has higher NDF digestibility and supports greater milk production and feed conversion efficiency than conventional hybrids (Oba and Allen, 1999b; Kung *et al.*, 2008). The *sfe* maize mutants with reduced ferulate lignin–arabinoxylan cross linkage also have higher cell wall digestibility and intake than conventional hybrids, resulting in higher milk production (Jung *et al.*, 2011). Barrier and Argiller (1993) highlighted the lower yield and susceptibility to lodging of brown midrib hybrids and suggested that genetic variation could lead to the selection of brown midrib hybrids of high agronomic value. Lauer and Coors (1997) reviewed 18 agronomic and dairy cattle feeding trials comparing brown midrib and conventional maize hybrids. They concluded that although NDF was lower for *bm3* than for conventional maize (by an average of 2%) and milk output per tonne of crop was higher (by 4%), yield per acre was lower for the bm3 hybrids by 6% and milk per acre was reduced by 2%.

Table 4: Effect of stage of forage maize harvest on AD specific methane yield and on methane yield per hectare

	Harvest 1	 2	 3
Harvest (days after sowing)	97	122	151
Stage of grain maturity	Milk ripeness	Wax ripeness	Full ripeness
Dry matter (g/kg fresh weight)[1]	187	293	468
Volatile solids (g/kg fresh weight)[1]	178	278	452
Specific methane yield (litres CH_4/kg volatile solids)[1]	338	308	278
Methane yield per hectare (m^3 CH_4)[2,3]	6350	7270	7930

[1] Means of three years and four late-maturing cultivars (FAO 290 to FAO 600).
[2] Means of three years and three late-maturing cultivars.
[3] m^3 = cubic metres at normal temperature and pressure.
Source: Amon *et al*., 2007b.

A very important attribute of maize for biogas is output per hectare of land, so yield of biomass (as DM or volatile solids) may be an overriding criterion in selection of species, cultivar and stage of plant maturity at harvest. Amon *et al.* (2007a) studied biomass and methane yields of a range of ensiled crops - maize, wheat, triticale, rye, sunflower and grass. They found that the highest methane yield per hectare was from maize harvested at the "wax ripeness" stage of maturity (300 to 350 g DM/kg fresh weight).

The effect of stage of maize plant maturity at harvest on specific methane yield and on methane yield per hectare is shown in Table 4. Specific methane yield decreased with advancing plant maturity. The decrease in specific methane yield with increased plant maturity reflected reduced concentration of fibre and increased concentration of starch in the whole plant DM, consistent with the reduction in methane production in the rumen with reduced proportion of acetate in the rumen VFA associated with increased concentrate in the diet (Yates, *et al*., 2000 and Table 1). However, despite reduced specific methane yield, the large increase in crop yield with advancing grain ripeness was reflected in an increase in methane output per hectare. Schittenhelm (2008) concluded that the ideal maize hybrid for biogas was a later-maturing hybrid that can be harvested at a DM concentration consistent with the production of good quality silage i.e. around 300 g DM/kg fresh weight.

Amon *et al.* (2004, 2007b) found that ensiling increased the specific methane yield of whole-crop maize by 0.25 compared to the fresh crop, presumably because the products of the silage fermentation were reduced compounds and more suitable substrates for utilisation by *Archaea* than the original water-soluble carbohydrate substrates. The possibility of directing fermentation in the silo by inoculating the crop at harvest was explored by Vervaeren *et al.* (2010) who added a range of inoculants to whole-plant maize ensiled at 26% DM. They found that specific methane yields after a 21-day incubation were higher from additive-treated than from untreated silage, and tended to be higher from silages treated with additives containing heterofermentative lactic acid bacteria (that produced lactic and acetic acids) together with cell wall degrading enzymes, than from silage treated with predominantly homofermentative lactic acid bacteria that produced lactic acid as the sole end product of fermentation (McDonald *et al*., 1991). The storage period may influence the efficacy of additive since Herrmann *et al*, (2011) found little effect of additive treatment on methane yield after an ensiling period of one year. They also noted a decrease in lactic acid and increases in acetic acid and in methane yield with increased length of storage period.

Several phenotypic characters of the maize plant have been found to exert a significant influence on methane production; namely crude protein, crude fat, cellulose and hemicellulose (Amon *et al*., 2004, 2007b). Calculation of theoretical biogas potential (gas yield and methane concentration) is possible from pre-determined concentrations of crude fibre, crude protein, crude fat, ash and moisture (Allison, 2011). Rath *et al.* (2013) found that concentrations of crude fat and hemicellulose in maize were positively related to biogas yield whilst acid detergent lignin and water-soluble carbohydrates were negatively related to biogas yield. In view of the positive relationship between crude fat concentration of maize and specific methane yield, cultivars with elevated concentrations of oil may be worth exploiting for AD, provided their biomass yield is competitive with conventional hybrids.

Grieder *et al.* (2012a, b, c) made a comprehensive study of genetic parameters of maize hybrids for biogas involving 570 testcross progenies of 285 inbred dent lines. Heritability estimates were high but genotypic variance and hence heritability in specific methane yield decreased towards the end of the 35-day fermentation, reflecting almost complete degradation of potentially digestible components during the relatively long residence period. Variation in total methane yield per hectare was mainly attributable to variation in DM yield. They concluded that introgression of later maturing or exotic material may be productive, with selection for higher DM yield and less focus on ear proportion for biogas maize compared to forage maize for animal feed.

In situations where large distances have to be travelled between field and farm, the cost of transportation of the crop is likely to be affected significantly by crop DM concentration. This cost should be taken into account in determining the optimal stage of plant maturity for harvesting for biogas production and supports full ripeness as the optimal stage of harvest for maximum methane yield per hectare (Table 4).

A challenge for the future is to optimise AD methane yield, ideally via in-line real-time analysis of feedstock composition using near infrared reflectance spectroscopy

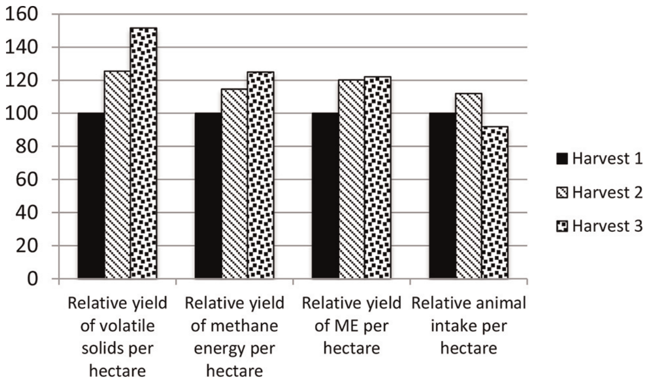

Figure 1: Effect of stage of maize crop harvest on relative yield of biomass, methane energy, ME and animal intake per hectare (Harvest 1 = 100). **Source:** Based on data in Table 4.

(Jacobi *et al.*, 2011), and also by determining factors in the ensiling process that impact significantly on methanogenesis.

In an attempt to integrate the effects of maize crop maturity on both biogas and animal nutrition, the data of Amon *et al*. (2007b, Table 4) and Oba and Allen (1999) were used to compare three dates of harvest in terms of biomass yield, methane energy yield, ME yield and animal intake per hectare of land. The results (Figure 1) are to be treated with caution since ME concentration and NDF digestibility were estimated for the purposes of the comparison. Nevertheless, the trends were similar for biomass, methane energy yield and ME yield, with yields increasing progressively with advancing crop maturity. Relative animal intake was highest at the medium crop maturity.

5. Conclusions

Yield of whole-plant maize biomass per hectare should be the main criterion of maize cultivar performance assessment for AD. Selection of cultivars for use in AD with elevated concentrations of oil or reduced concentrations of lignin may be desirable. Maize cultivars for use as animal feed should contain i) a relatively high proportion of ear in the total plant DM to give a high concentration of starch and ii) high NDF digestibility, to meet animal requirement for readily available rumen-fermentable forage energy. Selection of forage maize and other forage crop cultivars for both AD and animal feed should include evaluation of NDF concentration and NDF digestibility.

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REFERENCES

AL Seadi, T., Rutz, D., Prassl, H., Köttner, M., Finsterwalder, T., Volk, S. and Janssen, R. (2008). *Biogas Handbook*. Esbjerg, Denmark, University of Southern Denmark.

Allison, P. 2011. *The Calculation of Theoretical Biogas Potential*. Laboratory Standard Operating Procedure Manual SOP S1104, Sciantec Analytical Services Ltd, Cawood, UK.

Amon, T., Kryvoruchko, V., Amon, B., Zollitsch, W. and Pötsch, E. (2004). Biogas production from maize and clover grass estimated with the methane energy value system. In: *AgEng2004*. *Engineering the Future*, Leuven, Belgium, 12-16 September 2004.

Amon, T., Amon, B., Kryvoruchko, V., Machmüller, A., Hopfner-Sixt, K., Bodiroza, V., Hrbek, R., Friedle, J., Pötsch, E., Wagentristl, H., Schreiner, M. and Zollitsch, W. (2007a). Methane production through anaerobic digestion of various crops grown in sustainable crop rotations. *Bioresource Technology*, **98**, 3204-3212. DOI: 10.1016/j.biortech.2006.07.007

Amon, T., Amon, B., Kryvoruchko, V., Zollitsch, W., Mayer, K. and Gruber, L. (2007b). Biogas production from maize and dairy cattle manure – influence of biomass composition on the methane yield. *Agriculture, Ecosystems and Environment*, **118**, 173-182. DOI: 10.1016/j.agee.2006.05.007

Bal, M.A., Coors, J.G. and Shaver, R.D. (1997). Impact of the maturity of corn for use as silage in the diets of cows on intake, digestion and milk production. *Journal of Dairy Science*, **80**, 2497-2503. DOI: 10.3168/jds.S0022-0302(97)76202-7

Barrière, Y., Argillier, O., Michalet-Doreau, B., Hébert, Y., Guingo, E., Giauffret, C. and Émile, J.C. (1997). Relevant traits, genetic variation and breeding strategies in early silage maize. *Agronomie*, **17**, 395-411. DOI: 10.1051/agro:19970804.

Blaxter, K.L. and Clapperton, J.L. (1965). Prediction of the amount of methane produced by ruminants. *British Journal of Nutrition*, **19**, 511-522. DOI: http://dx.doi.org/10.1079/BJN19650046.

Blaxter, K.L. and Czerkawski, J. (1966). Modifications of the methane production of the sheep by supplementation of its diet. *Journal of the Science of Food and Agriculture*, **17**, 417-421. DOI: 10.1002/jsfa.2740170906.

Browne, I., Allen, D., Phipps, R.H. and Sutton J.D. (1995) *Mixed Forage Diets for Dairy Cows*. UK Milk Development Council.

Chamberlain, A.T. and Wilkinson, J.M. (1996). *Feeding the Dairy Cow*. Lincoln, UK, Chalcombe Publications.

Cherney, J.H., Cherney, D.J.R., Akin, D.E. and Axtell, J.D. (1991). Potential of brown-midrib, low-lignin mutants for improving forage quality. *Advances in Agronomy*, **46**, 157–198. doi: 10.1016/S0065-2113(08)60580-5.

DairyCo (2012). *Grassland Management Improvement Programme Third Edition*. DairyCo Grass+, Stoneleigh, UK, Agricultural and Horticultural Development Board.

DairyCo (2014). *Evidence Report*. DairyCo Milkbench+, Stoneleigh, UK, Agricultural and Horticultural Development Board.

Frei, M. (2013). Lignin: Characterization of a multifaceted crop component. *The Scientific World Journal*. http://www.hindawi.com/journals/tswj/2013/436517/ Accessed 7 February 2015. doi: 10.1155/2013/436517.

Garnsworthy P.C, Craigon J., Hernandez-Medrano J.H. and Saunders N. (2012). On-farm methane measurements during milking correlate with total methane production by individual dairy cows. *Journal of Dairy Science*, **95**, 3166-3180. doi: 10.3168/jds.2011-4605.

Giger-Reverdin, S., Morand-Fehr, P. and Tran, G. (2003). Literature survey of the influence of dietary fat composition on methane production in dairy cattle. *Livestock Production Science*, **82**, 73-79. doi: 10.1016/S0301-6226(03)00002-2.

Grieder, C., Mittweg, G., Dhillon, B.S., Montes, J.M., Orsini, E. and Melchinger, A.E. (2012a). Kinetics of methane fermentation yield in biogas reactors: Genetic variation and association with chemical composition in maize. *Biomass and Bioenergy*, **37**, 132-141. DOI: 10.1016/j.biombioe.2011.12.020.

Grieder, C., Dhillon, B.S., Schipprack, W. and Melchinger, A.E. (2012b). Breeding maize as biogas substrate in Central Europe: I. Quantitative-genetic parameters for testcross performance. *Theoretical and Applied Genetics*, **124**, 971-980. DOI: 10.1007/s00122-011-1761-y.

Grieder, C., Dhillon, B.S., Schipprack, W. and Melchinger, A.E. (2012c). Breeding maize as biogas substrate in Central Europe: II. Quantitative-genetic parameters for inbred lines and correlations with testcross performance. *Theoretical and Applied Genetics*, **124**, 981-988. DOI: 10.1007/s00122-011-1762-x.

Herrmann, C., Heiermann, M. and Idler, C. (2011). Effects of ensiling, silage additives and storage period on methane formation of biogas crops. *Bioresource Technology*, 102, n. 8, p. 5153-5161. DOI: 10.1016/j.biortech.2011.01.012.

Holter J.A. and Young A.J. (1992) Methane prediction in dry and lactating Holstein cows. *Journal of Dairy Science*, **75**, 2165-2175. DOI: 10.3168/jds.S0022-0302(92)77976-4.

Jacobi, H.F., Moschner, C.R. and Hartung, E. (2011). Use of near infrared spectroscopy in online monitoring of feeding substrate quality in anaerobic digestion. *Bioresource Technology*, **102**, 4688-4696. DOI: 10.1016/j.biortech.2011.01.035.

Johnson, K.A. and Johnson, D.E. (1995). Methane emissions from cattle. *Journal of Animal Science*, **73**, 2483-2492. DOI:/1995.7382483x.

Jung, H.G., Mertens, D.R. and Phillips, R.L. (2011). Effect of reduced ferulate-mediated lignin/arabinoxylan cross linking in corn silage on feed intake, digestibility and milk production. *Journal of Dairy Science*, **94**, 5124-5137. DOI: 10.3168/jds.2011-4495.

Lauer, J. and Coors, J. (1997). Brown midrib corn. *Corn Agronomy*, March 1997. http://corn.agronomy.wisc.edu/AA/A011.aspx. Accessed 12 May 2015.

Kleen J.L, Hooijer G.A., Rehage J. and Noordhuizen J.P.T.M. (2003). Subacute ruminal acidosis (SARA): a review. *Journal of Veterinary Medicine Series A* **50**, 406-414. DOI: 10.1046/j.1439-0442.2003.00569.x.

Kung, L. Jr, Moulder, B.M., Mulrooney, C.M., Teller, R.S. and Schmidt, R.J. (2008). Effect of silage cutting height on the nutritive value of a normal corn silage hybrid compared with brown midrib corn silage fed to lactating cows. *Journal of Dairy Science*, **91**, 1451-1457. DOI: 10.3168/jds.2007-0236.

McDonald, P., Henderson, A.R. and Heron, S.J.E. (1991). *The Biochemistry of Silage, Second Edition*, Marlow, UK, Chalcombe Publications.

McDonald, P., Edwards, R.A., Greenhalgh, J.F.D. and Morgan, C.A. (1995). *Animal Nutrition, Fifth Edition*, Harlow, UK, Longman.

McEvoy, T.G., Robinson, J.J., Aitken, R.P., Findlay, P.A. and Robertson, I.S. (1997). Dietary excess of urea influence the viability and metabolism of preimplantation sheep embryos and may affect fetal growth among survivors. *Animal Reproduction Science*, **47**, 71-90. DOI: 10.1016/S0378-4320(96)01627-2.

Mills, J.A.N., Crompton, L.A. and Reynolds, C.K. (2008). *Ruminant nutrition regimes to reduce methane and nitrogen emissions – a meta-analysis of current databases*. Report MDC/07/04A, Animal Science Research Group, University of Reading, UK.

Moss, A.R. 1993. *Methane – Global Warming and Production by Animals*. Canterbury, UK, Chalcombe Publications.

National Institute of Agricultural Botany, NIAB (2013) News: BSPB Launches Biogas Maize Descriptive List 04 July 2013. http://www.niab.com/news_and_events/article/286 Accessed 25 November 2014.

NIAB (2015) *2015 Forage Maize Descriptive List* http://www.niab.com/pages/id/332/Forage_Maize_Descriptive_List Accessed 9 February 2015.

Oba, M. and Allen, M.S. (1999a). Evaluation of the importance of the digestibility of neutral detergent fiber from forage: Effects on dry matter intake and milk yield of dairy cows. *Journal of Dairy Science*, **82**, 589-596. DOI: 10.3168/jds.S0022-0302(99)75271-9.

Oba, M., and Allen, M.S. (1999b). Effects of brown midrib 3 mutation in corn silage on dry matter intake and productivity of high yielding dairy cows. *Journal of Dairy Science*, **82**, 135-142. DOI: 10.3168/jds.S0022-0302(99)75217-3.

Offer, N.W., Givens, D.I., Blake, J.S. and Thomas, C. (2004). Decision support system to aid the process of ration formulation. In *Feed Into Milk: A New Applied Feeding System for Dairy Cows* (Ed C. Thomas), pp. 41-57. Nottingham, Nottingham University Press.

Opio, C., Gerber, P., Mottet, A., Falcucci, A., Tempio, G., Macleod, M., Vellinga, T., Henderson, B. and Steinfeld, H. (2013). Greenhouse gas emissions from ruminant supply chains – A global life cycle assessment. Rome, Food and Agriculture Organization of the United Nations.

Ørskov, E.R. (1998). *The Feeding of Ruminants. Second Edition*. Lincoln, UK, Chalcombe Publications.

Penner, G.B. (2014). Mechanisms of volatile fatty acid absorption and metabolism and maintenance of a stable rumen environment. *25th Florida Ruminant Nutrition Symposium,* 4 to 5 February 2014, pp. 92-104. http://dairy.ifas.ufl.edu/rns/2014/penner.pdf. Accessed 10 November 2014.

Rath, J., Heuwinkel, H. and Hermann, A. (2013). Specific biogas yield of maize can be predicted by the interaction of four biochemical constituents. *Bioenergy Research*, **6**, 939-952. DOI: 10.1007/s12155-013-9318-3.

Redman, G. (2010). *A Detailed Economic Assessment of Anaerobic Digestion Technology and its Suitability to UK Farming and Waste Systems. 2nd Edition.* Project NNFCC 10-010. 127pp. Melton Mowbray, UK, Andersons Centre.

Russell, J.B and Strobel, H.J. (1987). Effects of additives on in vitro ruminal fermentation: A comparison of monensin and bacitracin, another gram positive antibiotic. *Journal of Animal Science*, 66, 552-558. DOI:10.2134/jas1988.662552x.

Satter, L.D. and Slyter, L.L. (1974). Effect of ammonia concentration on rumen microbial protein production in vitro. *British Journal of Nutrition*, **32**, 194-208. DOI: http://dx.doi.org/10.1079/BJN19740073.

Schittenhelm, S. (2008). Chemical composition and methane yield of maize hybrids. *European Journal of Agronomy*, **29**, 72-79. DOI: 10.1016/j.eje.2008.04.001.

Schulze, A.K., Weisbjerg, M.R. and Nørgaard, P. (2014). Effect of level and NDF content of grass-clover silages on chewing activity, fecal particle size and NDF digestibility in dairy heifers. *Animal*, **12**, 1945-1954. DOI: 10.1017/S1751731114002055.

Strick, D.P.B.T.B., Domnanovich, A.M. and Holubar, P. (2006). A pH-based control of ammonia in biogas during anaerobic digestion of artificial pig manure and maize silage. *Process Biochemistry*, **41**, 1235-1238. DOI: 10.1016/j.procbio.2005.12.008.

Tamminga, S., Bannink, A., Dijkstra, J. and Zom, R. (2007). *Feeding strategies to reduce methane loss in cattle*. Report 34, Animal Sciences Group, Wageningen UR, The Netherlands.

Thomas, C. (Ed.) (2004). *Feed into Milk. A New Applied Feeding System for Dairy Cows. Feed Database*. Nottingham University Press, Nottingham, UK.

Van Soest, P.J. (1994). *Nutritional Ecology of the Ruminant, Second Edition*. Ithaca, USA, Cornell University Press.

Vellinga, T.V. and Hoving, I.E. (2011). Maize silage for dairy cows: mitigation of methane emissions can be offset by land use change. *Nutrient Cycling in Agroecosystems,* **89**, 413-426. DOI: 10.1007/s10705-010-9405-1.

Vervaeren, H., Ghekiere, G. and Willems, B. (2010). Biological ensilage additives as pretreatment for maize to increase the biogas production. *Renewable Energy*, **35**, 2089-2093. DOI: 10.1016/j.renene.2010.02.010.

Walker, N. (2014). They are what we feed them: Understanding the impact of forage quality on rumen function. *2014 Penn State Dairy Cattle Nutrition Workshop*, 12-13 November, Grantville, USA. http://extension.psu.edu/animals/dairy/courses/dairy-cattle-nutrition-workshop/previous-workshops/2014/breakouts-and-special-sessions/they-are-what-we-feed-them. Accessed 11 May 2015.

Weiland, P. (2010). Biogas production: Current state and perspectives. *Applied Microbiology and Biotechnology*, **85**, 849-860. DOI: 10.1007/s00253-009-2246-7.

Weissbach, F., Schmidt, F. and Hein, E. (1974). Method of anticipation of the run of fermentation in silage making, based on the composition of green fodder. *Proceedings of the XII International Grassland Congress, Moscow*, **III**, 663-673.

Weissbach, F. (2009). Prediction of biogas potential of silages. In: *Proceedings of the XV International Silage Conference,* (Eds. Broderick G.A., Adesogan A.C., Bocher L.W., Bolsen K.K., Contreras-Govea F.E., Harrison J.H. and Muck R.E.), pp. 189-190. Madison, Wisconsin, USA.

Wilkinson, J.M. (2005) *Silage*. Lincoln, UK, Chalcombe Publications.

Wilkinson, J.M. (2013). Trends and targets in milk production from forage. In: *Profitable and Sustainable Grazing Systems – Moving Forward with Science.* (Eds Moorby J.M. and Mayne C.S.) Proceedings BGS/BSAS Conference, Malvern, 26 and 27 February 2013. p. 81.

Wilkinson, J.M. and Phipps, R.H. (1979). The development of plant components and their effects on the composition of fresh and ensiled forage maize. 2. The effect of genotype, plant density and date of harvest on the composition of maize silage. *Journal of agricultural Science, Cambridge,* **92**, 485-491. DOI: http://dx/doi.org/10.1017/S0021859600063024.

Wilkinson, J.M., Newman, G. and Allen, D.M. (1998). *Maize: Producing and Feeding Maize Silage*. Lincoln, UK, Chalcombe Publications.

Yates, C.M., Cammell, S.B., France, J. and Beever, D.E. (2000). Prediction of methane emissions from dairy cows using multiple regression analysis. *Proceedings of the British Society of Animal Science, 2000*, p. 94.