*Indian Journal of Animal Sciences* **84** (3): 306–310, March 2014/Article

# Silages prepared from different maize hybrids effect *in-vitro* rumen fermentation parameters and growth performance in crossbred calves

SHWETA SINGH[1], AMRISH KUMAR TYAGI[2] and VISHNU KALE[3]

*National Dairy Research Institute, Karnal, Haryana 13200 India*

Received: 30 June 2013; Accepted: 30 November 2013

ABSTRACT

The experiment was conducted to evaluate the effects of silages prepared from different maize varieties/hybrids on fermentation characteristics, microbiological characteristics, and growth performance in growing crossbred calves. Female crossbred calves (18) of 4–5.5 months of age were selected and distributed randomly into 3 treatment groups of 6 animals each, based on their body weight. All the calves of 3 groups were offered silages prepared from different varieties/hybrids (control - Vijay composite, T1 - DKC 9108 and T2 - 900M Gold) and concentrate in 70: 30 ratio in 105 days of experimental period. Metabolism trial was conducted to assess feed intake, nutrients utilization and N balance. *In vitro* gas/methane production and digestibility trials were carried out for 24 h and 48 h respectively. *In vitro* methane production was higher in T1 than control and T2. The total DMI (kg/day) as well as body weight change in crossbred calves was similar in 3 groups throughout the experimental period. TDNI (kg/day) and NDF digestibility was significantly higher in T1 than control and T2 groups. CP digestibility was higher in control and T1 than T2. The results showed that DMI and growth performance of calves were similar in 3 groups notwithstanding the different chemical composition as well as different microbial characteristics of silages prepared from different maize varieties/hybrids. However methane production differed significantly among the groups which can be an area of interest for further mitigation studies.

*Abbreviations:* DMI: Dry matter intake, IVFA, individual volatile fatty acids, IVGP: *in vitro* gas production, IVMP: *in vitro* methane production, IVTDMD: *in vitro* true dry matter digestibility, IVTOMD: *in vitro* true prganic matter digestibility, IVNDFD: *in vitro* neutral detergent fiber digestibility, NDF : neutral detergent fibre, NDS : neutral detergent solution, $NH_3$-N : ammonia-N, TDNI: total digestible nitrogen intake, TVFA: total volatile fatty acid.

**Key words**: Crossbred calves, Growth performance, Maize varieties/hybrids, Silage

The current annual production of green and dry fodder in the country is 400.6 and 466 million tonnes as against estimated requirement of 1,097 and 609 million tonnes. The shortage of dry fodder, concentrate and green fodder is 10, 33 and 35% respectively. Only 25% of forage seeds are available that too of 15–20 years old varieties. Fodder is being cultivated in only about 4% of the agricultural land, and is not adequate to meet the requirement of fodder in the country (Planning Commission of India 2012). Any good forage crop should have high dry matter yield, high protein content, high energy content (high digestibility), high intake (low fibre) and optimum dry matter content at harvest for acceptable fermentation and storage. With the exception of high protein level, maize fodder exhibits these characteristics better than other forages. Maize is easily ensiled and results in palatable forage with relatively consistent quality and higher energy content than other forages. Both hybrid selection and agronomic management influence silage yield and quality (Lauer 2013). Seed companies are developing hybrids that are targeted to silage producers (Ballard *et al*. 2001). Leafy, waxy and brown midrib hybrid types are being developed specifically for silage production (Cox and Cherney 2001).

Thus, in the present investigation efforts were made to compare the conventional variety silage and corn hybrid silage and to determine the impact of corn hybrid silage on *in vitro* rumen fermentation parameters and the performance of growing crossbred calves.

## MATERIALS AND METHODS

Maize variety Vijay composite, Maize hybrid DKC 9108 and Maize hybrid 900 M Gold, were planted on the National Dairy Research Institute's farm with seed rate 25 kg/acre and harvested at soft dough stage after 70 days of sowing. Chaffed green fodders were preserved in tower silo (capacity 2.53 cubic metre). Molasses @ 2% and salt @ 0.5% were mixed with fodder, packed air tightly by double polythene sheets and stored for 60 days.

Present address: [1]Post-graduate student, [2]Principal Scientist (amrishtyagi1963@yahoo.com), [3]Ph D Student, Dairy Cattle Nutrition Division.

*Analysing silage for quality:* At the end of 60 days storage period, the polythene sheets were opened. Water extract of silage was prepared by adding 100 ml of distilled water to 10 g of fresh samples in a beaker and homogenized by mechanical homogenizer. The material was filtered through four layers of cheese cloth and stored in refrigerator at 4°C. The pH of water extract was estimated by digital pH meter. The fresh silage samples were analysed for dry matter (Dewar and McDonald 1961) by using Dean and Stark water estimation trap with Liebig condenser. The estimation of lactic acid in silage samples was done as per Barker and Summerson (1941) and modified by Barnett (1951). Total nitrogen and $NH_3$-N content was estimated in fresh silage samples by micro Kjeldahl method.

*Microbial analysis:* For preparation of silage extract, silage (20 g) was added to 180 ml of saline (0.85% sodium chloride in distilled water) and mixed vigorously for 4–5 min. The suspension was serially diluted ($10^3$ to $10^8$) and 1 ml aliquots from each dilution were spread in duplicate onto *Lactobacilli* agar media (Rogosa agar medium) and Coliform agar media (EMB agar media) for enumeration of *Lactobacilli* and *Coliform*, respectively. Rogosa agar plate and EMB agar plates were incubated at 37°C for 24 h. Colonies were counted from plates containing a minimum of 30 and maximum of 300 colonies.

In vitro *study*: *In vitro* trials were conducted in triplicate to estimate the various parameters such as total gas production, methane production and *in vitro* DM/NDF digestibility. Rumen liquor was collected from a fistulated male cattle maintained on a standard diet (60 parts roughage: 40 parts concentrate) before morning feeding and watering into a pre-warmed thermos flask. The samples were incubated with rumen fluid in calibrated glass syringes (Menke and Steingass 1988). The sample (0.200 g dry weight) was weighed on a plastic boat and was placed into the bottom of the glass. The syringes were pre-warmed at 39°C before the injection of 30 ml rumen fluid-buffer mixture into each syringe followed by incubation in water-bath at 39°C. The syringes were gently shaken 30 min after the start of incubation and every hour for the first 10 h of incubation. Gas production was recorded 24 h after incubation. Proportion of methane in total gas was measured using gas liquid chromatograph fitted with flame ionization detector (FID) and a column. Gas sample from the syringe (5 ml) was injected into the GLC through injection port. In the same manner, a standard having 50% $CH_4$ and 50% $CO_2$ was injected for comparison. The temperature of injector, detector and oven of gas chromatograph was maintained as 140, 200 and 250°C, respectively, and the flow rate for carrier gas (N), air through FID and fuel gas (IOLAR grade hydrogen) and was 30, 300 and 30 ml/min, respectively. Samples of silages were subjected to a 48 h digestion period with rumen fluid-buffer mixture in glass syringes for determination of true DM and NDF digestibility (Goering and Van Soest 1970). The estimated ME value (MJ/kg DM) was calculated using the equations of Menke and Steingass (1988) as follows:

$$\text{ME (MJ/kg DM)} = 2.20 + 0.136{*}\text{GP} + 0.057{*}\text{CP} + 0.0029{*}(\text{CP})^2$$

*Management of animals and dietary treatments:* Eighteen female crossbred (KF) growing calves of 4–5.5 months of age with average body weight of 60 kg were selected from the cattle yard of the institute and distributed randomly into 3 treatment groups of 6 animals each based on their body weight and age. All the animals were housed in the experimental sheds of the institute, which were well ventilated and having individual pens to facilitate individual feeding. Standard prophylactic measures including deworming and spraying of insecticides was followed during the experimentation. The calves under treatment group 1 (T1) were fed 70% silage (corn hybrid DKC 9108) + 30% concentrate and treatment group 2(T2) were fed 70% silage (corn hybrid 900 M Gold) + 30% concentrate while the calves under control (C) group were fed 70% silage (Vijay composite) + 30% concentrate. Ingredient composition of concentrate mixture (% DM basis) was 33% maize grain, 18% GNC exp., 10% MOC exp., 6% DORB, 5% *gaur* meal, 20% wheat bran, 5% pearl millet, 2% mineral mixture and 1% salt.

Nutrient requirement were fulfilled by NRC (2001) feeding standard. For determining the nitrogen balances, digestibility of DM, nutrient intake and nutrient utilization 6-day metabolism trial was conducted at the end of growth experiment.

*Sampling and analysis of feed, faeces and urine:* Dry matter, organic matter, crude protein, ether extract and total ash in feeds and faeces samples were determined (AOAC 2005). The fractions of cell wall constituents (NDF, ADF and ADL) were also determined (Van Soest *et al.* 1991). Urine samples were analyzed for nitrogen content (AOAC 2005).

*Statistical analysis:* The results obtained during this study were analyzed by using software package IBM SPSS version 21. Tueky multiple range test was used to differentiate the means at ($P < 0.05$) level of significance.

## RESULTS AND DISCUSSION

*Fermentation and microbiological characteristics of silages prepared from different maize varieties/hybrids:* The data on fermentation and microbiological characteristics of silages prepared from different maize varieties/hybrids in control and treatment groups (Table 1) indicated that silage pH ranged from 4–4.2, which is also reported by Kung and Stoke (2001). Lactic acid contents varied from 5.22 to 7.20%. Lactic acid is the most abundant acid (75% of the total acids contained in silage) and also stronger than the volatile fatty acids (VFA) and therefore has the greatest effect on silage pH (Kung 2001, Seglar 2003). It ranged between 4 and 7% for maize silage (Seglar 2003). Lactic acid in T1 group was of highest concentration, directly correlating with the results obtained in microbial analysis i.e. high number of lactobacillus count. There was wide variation in the TVFA content (mM/100 g DM). It was

Table 1. Fermentation and microbiological characteristics of silages prepared from different maize varieties/hybrids

| Parameters | Control | T1 | T2 |
|---|---|---|---|
| pH | 4.20 | 4.03 | 4.21 |
| Lactic acid (g/100g DM) | 6.51±0.66 | 7.20±0.74 | 5.22±0.69 |
| TVFA (mM / 100g DM) | 55.76±2.55 | 37.22±1.81 | 35.15±1.54 |
| Acetate (% DM) | 2.70±0.11 | 1.92±0.09 | 1.85±0.09 |
| Propionate (% DM) | 0.68±0.07 | 0.33±0.02 | 0.28±0.02 |
| Butyrate (% DM) | 0.12±0.01 | 0.06±0.005 | 0.04±0.004 |
| Total-N (g/100 g DM) | 2.32±0.01 | 2.13±0.02 | 2.17±0.05 |
| $NH_3$-N (g/100 g DM) | 0.18±0.01 | 0.10±0.01 | 0.11±0.02 |
| Lactobacillus (cfu/g) | $1.80^b\pm 0.60\times10^9$ | $8.70.14^c \pm0.11\times10^9$ | $3.47^a \pm0.65\times10^7$ |
| Coliform (cfu/g) | $1.76^b$ ±0.8×104 | $0^a$ | $3.24^c$ ±0.5×104 |

Table 2. Chemical composition (% DM) of silages prepared from different maize varieties/hybrids

| Parameters | Control | T1 | T2 |
|---|---|---|---|
| DM | 26.84±0.115 | 28.94±0.05 | 32.41±0.05 |
| OM | 85.93±0.04 | 83.57±0.05 | 83.72±0.15 |
| CP | 14.57±0.01 | 13.34±0.02 | 13.65±0.03 |
| EE | 2.53±0.01 | 2.36±0.01 | 3.44±0.01 |
| NDF | 60.32±0.01 | 62.10±0.01 | 60.37±0.05 |
| ADF | 36.38±0.07 | 33.04±0.02 | 36.42±0.01 |
| ADL | 3.35±0.01 | 2.58±0.10 | 3.68±0.05 |
| TDN (%) | 60.88±0.05 | 63.20±0.05 | 60.52±0.05 |
| ME(MJ/kg DM) | 9.21±0.07 | 9.56±0.02 | 9.15±0.01 |

highest in control and lowest in T2. The VFA that has the biggest impact on aerobic stability is acetic acid, which was up to 3% concentrations (Danner *et al.* 2003, Muck 2010). Seglar (2003) also observed that propionic acid in well fermented maize silage was less than 0.5%, with butyric acid being undetectable (< 0.1%). This, however, was not the case for control in this study. The propionic acid and butyric acid concentrations (0.68 and 0.12% respectively) of the silage were slightly higher than expected. Kung and Shaver (2001) reported that high levels of silage acids indicate the occurrence of extensive fermentation in the silo. Wet silages that have undergone a long fermentation sometimes contain higher levels of acetic acid. The excessive fermentation due to high moisture (73.16%) may be a reason for the high volatile fatty acid concentration in control.

There was not much variation in nitrogen fractions (% DM) of different silages. Sheperd and Kung (1996) postulated that $NH_3$-N concentration of corn silage ranged between 0.04 and 0.15% of DM. The ammonia-N content of the T1 (0.10±0.01) and T2 (0.11±0.02) in the present study fell within the range except for the control (0.18±0.01) which was slightly at the upper limits of what can be expected for maize silage. *Lactobacillus* count was significantly higher in T1 ($8.70.14^c\pm0.11\times10^9$) than control ($1.80^b\pm0.60\times10^9$) and T2 ($3.47^a\pm0.65\times10^7$). When the *Lactobacilli* reach a level of at least $10^5$ cfu/g of silage, silage can be well preserved (Addah *et al*. 2011). Coliform bacteria was absent in TI. Our results are in line with that of Sanderson *et al.* (2005) who observed that coliform counts ranged from 0 to $1.24\times10^7$ with a median of 6,800 CFU / gram corn silage.

*Chemical composition of maize silages:* Detailed chemical composition (% DM) of maize silages prepared from different maize varieties/hybrids is given in Table 2. The DM content varied from 26.84±0.115 to 32.41±0.05. Control group recorded higher CP content than T1 and T2. There was no variation in the NDF content of different silages. CP values of silages in the present study were higher than NRC (2007) values. However, Tjardes and Allen (2000) and Hamaza *et al.* (2009) reported 12.9 and 14.05% CP for whole corn silage and *bmr* corn hybrid silage, respectively. Hutjens (1998) tested 86 varieties of corn silage and found that NDF and ADF ranged between 41.2 and 70.9 and 21.7 and 40.7%, respectively.

In vitro *study*: Data pertaining to *in vitro* study are presented in Table 3. IVDMD of T1 ($75.36^b\pm0.48$) was higher than that of control ($72.03^a\pm0.78$) and T2 (70.18

Table 3. *In vitro* digestibility (incubation period 48 h), *in vitro* gas production and rumen fermentation parameters (incubation period 24 h)

| Parameters | Control | T1 | T2 |
|---|---|---|---|
| IVDMD | $72.03^a$ ±0.78 | $75.36^b$ ±0.48 | $70.18^a$ ±0.15 |
| IVNDFD | $53.64^b$ ±0.86 | $60.33^c$ ±1.15 | $50.61^a$ ±0.8 |
| *Nitrogen fractions (mg/litre)* | | | |
| Total N | $980.61^c$ ±0.20 | $770.47^a$ ±0.58 | $840.32^b$ ±0.33 |
| TCA precipitable N | $541.25^c$ ±0.27 | $419.26^a$ ±0.53 | $477.96^b$ ±0.31 |
| $NH_3$-N | $231.11^c$ ±0.36 | $224.07^b$ ±0.45 | $213.31^a$ ±0.70 |
| *Fractions of VFA (mmol/litre)* | | | |
| TVFA concentration | 56.83 ±2.03 | 62.49 ±0.52 | 60.45 ±1.08 |
| Acetate concentration | $32.44^a$ ±0.70 | $35.08^b$ ±0.52 | $33.81^{ab}$ ±0.58 |
| Propionate concentration | 15.54 ±0.88 | 16.85 ±0.55 | 16.08 ±0.25 |
| Butyrate concentration | 8.45 ±0.51 | 10.11 ±0.60 | 10.15 ±0.48 |
| Total gas (ml/g DM) | $112.47^a$ ±0.74 | $146.33^b$ ±0.56 | $144.64^b$ ±0.52 |
| Methane as % of total gas | $17.03^a$ ±0.75 | $23.39^b$ ±0.32 | $16.74^a$ ±0.67 |
| Methane (ml/g DM) | $19.15^a$ ±0.78 | $34.23^c$ ±0.54 | $24.22^b$ ±1.02 |
| ME (MJ/kg DM) | $6.69^a$ ±0.02 | $7.50^b$ ±0.01 | $7.41^b$ ±0.01 |

Figures bearing different superscripts [a,b] within a column and in overall means differ significantly (P<0.05).

Table 4. DMI and body weight changes in cross bred calves fed on ration containing silages prepared from different maize varieties/hybrids throughout the experimental period

| Parameters | Control | T1 | T2 |
|---|---|---|---|
| DMI (kg/day) | 2.87±0.05 | 3.08±0.10 | 2.95±0.06 |
| DMI (kg/100 kg B.W.) | 3.00±0.07 | 3.00±0.26 | 3.20±0.26 |
| DMI(g/kg $w^{0.75}$) | 94.31±5.01 | 96.03±4.13 | 99.66±5.06 |
| Initial body weight (kg) | 60.25±5.84 | 59.90±6.57 | 60.36±6.03 |
| Final body weight (kg) | 95.41$^{ab}$±4.26 | 102.38$^{a}$±5.44 | 92.07$^{b}$±3.64 |
| Weight gain (kg) | 35.16$^{ab}$±0.23 | 42.48$^{a}$±0.23 | 31.71$^{b}$±0.32 |
| Daily weight gain (kg/day) | 0.334$^{ab}$±0.01 | 0.404$^{a}$±0.01 | 0.302$^{b}$±0.01 |
| FCR(kg gain/kg feed DMI) | 0.11$^{a}$±0.01 | 0.13$^{b}$±0.01 | 0.10$^{a}$±0.01 |

Figures bearing different superscripts $^{a,b}$ within a column and in overall means differ significantly (P<0.05).

$^{a}$±0.15) but no difference was observed between control and T2. IVNDFD of T1 (60.33$^{c}$±1.15) was higher than control (53.64$^{b}$±0.86) and T2 (50.61$^{a}$±0.8). The IVTGP from control (112.47 $^{a}$±0.74) was lower than T1 (146.33$^{b}$±0.56) and T2 (144.64 $^{b}$±0.52) and no difference was observed between T1 and T2. *In vitro* methane production (IVMP%) was higher in T1 (23.39$^{b}$±0.32) than control (17.03$^{a}$±0.75) and T2 (16.74$^{a}$±0.67). The results of the IVGP in the present study are in line with those of Calsamigila *et al*. (2007) and Calabro *et al*. (2007).

*DMI and body weight changes:* Data pertaining to DMI and body weight changes are presented in Table 4. The mean DMI kg/day, kg/100 kg body weight and g/kg $w^{0.75}$ were similar in the 3 groups. Final body weight, body weight gain and average daily gain were higher in T1 as compared to T2 when these were compared with control there was no difference. Our results are in line with several other studies that reported that feeding of corn hybrid silages did not affect DMI in Holstein cows (Taylor and Allen 2005, Weiss and Wyatt 2006). Our results pertaining to the body weight changes corroborated with other similar studies which have not found any significant difference in body weight gain in Holstein cows (Castro *et al.* 2010, Holt *et al*. 2012).

*Metabolic study:* Data pertaining to chemical composition of experimental diets, nutrient intake and nutrient digestibility during metabolism trial are presented in Table 5. Diet of control group contained higher CP content than that of T1 and T2. CPI was similar among all the groups. TDNI (kg/day) was higher in T1 than control and T2 groups. The present findings for DMI and CPI are similar to that of Nennich *et al.* (2003) who also did not observe any significant difference in DMI and CPI in Holstein Fresian cows. DM and OM digestibility coefficient were similar in all the 3 groups. CP and ADF digestibilities were higher in control and T1 than T2. NDF digestibility was higher in T1 than control and T2. DM digestibilities are in agreement with those of Castro *et al*. (2010) who did not observe any significant difference between DM digestibility of corn variety and corn hybrid silage. Nennich *et al.* (2003) reported that CP digestibility were significantly different in Holstein Fresian cows fed on different corn hybrid silages. NDF digestibilities were in agreement with studies which have those observed higher NDF degradation rates in corn hybrid silage compared with the conventional variety silage (Taylor and Allen 2005, Weiss and Wyatt 2006). N intakes, total N excreted and N balance in all the 3 groups were similar.

Table 5. Chemical composition, Nutrient intake, nutrient digestibility and N- balance of crossbred calves fed on ration containing silages prepared from different maize varieties/ hybrids during metabolic trial

| Nutrients | Control | T1 | T2 |
|---|---|---|---|
| DM | 46.35±0.01 | 48.06±0.02 | 51.13±0.01 |
| OM | 87.79±0.02 | 86.11±0.02 | 86.25±0.03 |
| CP | 15.46±0.01 | 14.62±0.02 | 14.83±0.03 |
| EE | 3.04±0.03 | 2.90±0.03 | 3.67±0.01 |
| NDF | 51.54±0.02 | 52.90±0.03 | 51.53±0.03 |
| ADF | 28.63±0.03 | 26.41±0.01 | 28.91±0.02 |
| ADL | 3.80±0.02 | 3.32±0.04 | 4.08±0.05 |
| TDN | 66.32±0.1 | 67.41±0.2 | 66.06±0.2 |
| ME (MJ/kg DM) | 12.34±0.01 | 12.54±0.03 | 12.29±0.03 |
| Body weight (kg) | 80.82$^{a}$±4.31 | 92.48$^{b}$±5.41 | 77.97$^{a}$±3.90 |
| DMI (kg/day) | 2.84±0.41 | 3.05±0.72 | 2.91±0.54 |
| CP intake (kg/day) | 0.45±0.04 | 0.46±0.03 | 0.44±0.26 |
| TDNI (kg/day) | 1.78$^{a}$±0.27 | 1.96$^{b}$±0.49 | 1.82$^{a}$±0.35 |
| *Nutrient digestibility (%)* | | | |
| DM | 61.91±1.42 | 66.16±1.6 | 60.20±1.8 |
| OM | 64.58±1.45 | 68.97±2.17 | 63.05±2.15 |
| CP | 71.60$^{b}$±2.18 | 68.16$^{b}$±1.89 | 60.29$^{a}$±1.99 |
| NDF | 60.34$^{a}$±1.60 | 67.61$^{b}$±2.00 | 58.08$^{a}$±1.85 |
| *Nitrogen balance* | | | |
| ADF | 63.38 $^{b}$±2.26 | 66.57 $^{b}$±1.96 | 54.31$^{a}$±1.54 |
| N intake (g/d) | 72.75±1.97 | 73.94±1.84 | 71.07±1.87 |
| N voided in faeces (g/d) | 20.29$^{a}$±0.88 | 23.29$^{a}$±1.60 | 28.76$^{b}$±1.11 |
| N voided in urine (g/d) | 30.51$^{b}$±0.95 | 28.29$^{ab}$±1.02 | 26.27$^{a}$±3.10 |
| Total N outgo (g/d) | 50.80±1.00 | 51.58±1.50 | 55.04±1.25 |
| N balance (g/d) | 21.94±2.53 | 22.35±0.93 | 16.04±2.01 |

Figures bearing different superscripts $^{a,b}$ within a column and in overall means differ significantly (P<0.05).

From our results it can be concluded that silage quality of all the 3 maize varieties/hybrids was similar in terms of fermentation and microbiological characteristics. *In vitro* evaluation suggests that gas production, methane production, dry matter digestibility and NDF digestibility were higher in silage of DKC 9108. DMI and growth performance was similar among all the 3 groups.

## ACKNOWLEDGEMENT

The authors would like to place on records the assistance rendered by Director, Incharge, Forage Section and

Incharge, Livestock Research Farm, National Dairy research Institute, Karnal for providing necessary facilities for conducting the research.

REFERENCES

Addah W, Baah J, Groenewegen P, Okine E K and McAllister T A. 2011. Comparison of the fermentation characteristics, aerobic stability and nutritive value of barley and corn silages ensiled with or without a mixed bacterial inoculants. *Canadian Journal of Animal Science* **91**: 133–46.

AOAC. 2005. Official methods of analysis. *Association of Official Analytical Chemists.* 17th edn. Arlington, Virginia, 22201,USA.

Ballard C S and Thomas E D. 2001. Effect of corn silage hybrid on dry matter yield,nutrient composition, *in vitro* digestion, intake by dairy heifers, and milk production by dairy cows. *Journal of Dairy Science* **84**: 442–52.

Barker S B and Summerson W H. 1941. The colorimetric determination of lactic acid in biological materials. *Journal of Biological Chemistry* **138**: 535.

Barnett A J G. 1951. The colorimetric determination of lactic acid in silage. *Biochemical Journal* **49**: 527–29.

Calabrò S, Tudisco R, Grossi M, Gonzalez O J, Caiazzo C, Guglielmelli A, Piccolo V, Infascelli F and Cutrignelli M I. 2007. Nutritive value of silages utilized in buffalo nutrition. Dipartimento di Scienze Zootecniche e Ispezione degli alimenti, University of Naples Federico II, Via F. Delpino, 1 80137, Napoli (Italy).

Calsamiglia S, Hernandez B, Hartnell G F and Phipps R. 2007. Effects of corn silage derived from a genetically modified variety containing two transgenes on feed intake, milk production, and composition and the absence of detectable transgenic deoxyribonucleic acid in milk in holstein dairy cows. *Journal of Dairy Science* **90**: 4718–23.

Castro J J, Bernard J K, Mullis N A and Eggleston R B. 2010. Brown midrib corn silage and Tifton 85 bermuda grass in rations for early lactation cows. *Journal of Dairy Science* **93**: 2143–52.

Cox W J and Cherney D J R. 2001. Influence of brown midrib, leafy and transgenic hybrids on corn forage production. *Agronomy Journal* **93**: 790–96.

Danner H, Holzer M, Mayrhuber E and Braun R. 2003. Acetic acid increases stability of silage under aerobic conditions. *Applied and Environmental Microbiology* **69**(1): 562–67.

Dewar W A and McDonald P. 1961. Determination of dry matter in silage by distillation with toluene. *Journal of the Science of Food and Agriculture* **12**: 790–93.

Goering H K and Van Soest P J. 1970. Forage fibre analysis: Apparatus, reagents, procedures and some applications. *Agricultural Handbook*. No. 379. USDA-ARS, U.S. Department of Agriculture Washington DC, U.S.

Hamaza A S, Shinnawy M M, Emara M F, Mohammadi T F and Shinnawy A M. 2009. Use of different silages as new feed resources for ruminants. *Archiva Zootechnica* **12**: 2, 79–88.

Holt M S, Eun J S, Thacker C R, Young A J, Dai X and Nestor K E. 2012. Effects of feeding brown midrib corn silage with a high dietary concentration of alfalfa hay on lactational performance of Holstein dairy cows for the first 180 days of lactation. *Journal of Dairy Science* **96**: 515–23

Hutjens M. 1998. *Evaluating Corn Silage Digestibility.* Extension Dairy specialist, University of Illinois, US. 1–7.

Kung L. 2001. Silage fermentation and additives. *Animal Feed Science and Technology* **17**: 145–59.

Kung L and Shaver R. 2001. Interpretation and use of silage fermentation analysis reports. *Focus on Forage*. **3**(13), University of Wisconsin Extension.

Kung L and Stokes M. 2001. Analyzing silages for fermentation end products http: //www.ag.udel.edu/anfs/faculty/kung/articles/analyzing_silages_for_formentati.htm [2012, November 21].

Lauer J. 2013. *More Mileage From Corn Silage: Selecting Hybrids*. Agronomy Department, University of Wisconsin, Madison, U.S.

Menke K H and Steingass H. 1988. Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. *Animal Research* and *Development* 28: 7–55.

Muck R E. 2010. Silage additives and management issues. *Proceedings of Idaho Alfalfa and Forage Conference.*Best Western Burley Inn, Burley, Idaho, US. 1–17.

National Research Council (NRC). 2007. *Nutrient Requirements of Horses.* 6th revised edn. National Research Council, National Academy Press, Washington, DC.

National Research Council (NRC).2001. *Nutrient Requirements for Dairy Cattle*. 7th revised edn. National Research Council, National Academy Press, Washington, DC.

Nennich D, Linn J G, Johnson D G, Endres M I and Jung H G. 2003. Comparison of feeding corn silages from leafy or conventional corn hybrids to lactating dairy cows. *Journal of Dairy Science* **86**: 2932–39.

Planning commission of India. 2012–2017. *Report of the Working Group on Animal Husbandary and Dairying.* 12th Five Year Plan, New Delhi, India.

Sanderson M W, Sargeant J M, Renter D G, Griffin D D and Smith R A. 2005. Factors associated with the presence of coli forms in the feed and water of feedlot cattle. *Applied and Environmental Microbiology* **71**: 6026–32.

Seglar B. 2003. Fermentation analysis and silage quality testing. *Proceedings of the Minnesota Dairy Health Conference.* College of Veterinary Medicine, University of Minnesota, US. 119–136.

Sheperd A C and Kung L. 1996. An enzyme additive for corn silage: Effect on silage composition and animal performance. *Journal of Dairy Science* **79**: 1760.

Taylor C C and Allen M S. 2005. Corn grain endosperm type and brown midrib 3 corn silage: Ruminal fermentation and N partitioning in lactating cows. *Journal of Dairy Science* **88**: 1434–42.

Tjardes K E and Allen M S. 2000. Brown midrib 3 corn silage improves digestion but not performance of growing beef steers. *Journal of Animal Science* **78**: 2957–65.

Van Soest, P.J., Robertson, J.B. and Lewis, B.A. 1991. Methods for dietary fiber, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science* **74**(10): 3583–97.

Weiss W P and Wyatt D J. 2006. Effect of corn silage hybrid and metabolizable protein supply on nitrogen metabolism of lactating dairy cows. *Journal of Dairy Science* 89: 1644–53.