EFFECT OF ADDITION OF DIFFERENT LEVELS OF FORMIC ACID AND UREA ON CHEMICAL COMPOSITION AND FERMENTATION CHARACTERISTICS OF WILD REED *PHRAGMITIS COMMUNIS* SILAGE

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ABSTRACT

This study was carried out in the laboratory to investigate the effect of addition of different levels of formic acid (FA) and urea on chemical composition and fermentation characteristics of common reed silages. About 400-500g of silage samples were prepared by treating chopped reed plants (2-3 cm) with solutions containing 10% of date juice, 0.5, 1 or 1.5% of FM and 0, 1 or 2% of urea. Silage samples were packed in double nylon bags and stored anaerobically for 60 days. Results showed that green color was roughly dominant in most samples of silage with vinegar odor. Samples were well aggregated with little mold was observed in few urea untreated silages. Results revealed also that increasing level of FA from 0.5 to 1 and 1.5% increased ($P<0.01$) contents of dry matter (DM) by 0.79 and 1.15%, and crude protein (CP) by 1.42 and 2.11% respectively, and decreased ($P<0.01$) ether extract (EE), by 0.29 and 0.63%. About effect of urea levels, most variables pointed out that there was a decrease may be occurred in fermentations due to a significant decrease in contents of DM ($P<0.01$) and EE ($P<0.05$). Content of crude fiber (CF) was decreased ($P<0.01$) from 44.7 to 43 and 41.1% for 0, 1and 2% levels of urea respectively. Results showed that there was a decrease ($P<0.01$) in pH values from 5.90 to 4.99 and 4.88, concentrations of ammonia nitrogen (NH_3-N), from 1.19 to 0.75 and 0.66% of total nitrogen and total volatile fatty acids (TVFA) from 6.56 to 4.61 and 4.14 mmol/100 g DM of silage samples as a result of addition of FA at levels of 0.5, 1 and 1.5% respectively. However, increasing urea levels from 0 to 1 and 2% associated with an increase ($P<0.01$) in fermentation parameters, 5.02, 5.06 and 5.70 for pH, 0.67, 0.98 and 0.95 for NH_3-N and 3.70, 5.53 and 6.07 mmol/100 g DM for TVFA respectively

Key word: Reed, Silage, Formic acid, Urea, Fermentation

تأثير اضافة مستويات حامض الفورميك واليوريا في التركيب الكيميائي ومعايير تخمرات لسايلج القصب البري *Phragmitis communis*

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المستخلص

اجريت هذه الدراسة المختبرية للتحري عن تأثير اضافة مستويات مختلفة من حامض الفورميك واليوريا في تخمرات سايلج القصب البري وتركيبه الكيميائي. صنعت نماذج السايلج (400-500 غم) بمعاملة القصب المقطع الى 2-3 سم بمحلول الاضافة المحتوي على عصير التمر بمستوى 10% و ثلاث مستويات من اليوريا 0 و 1 او 2% وثلاث مستويات ايضا من حامض الفورميك 0.5 و 1 او 1.5%. عبئت النماذج في اكياس نايلون مزدوجة وحفظت لاهوائيا لمدة 60 يوم. اظهرت النتائج ان اللون الاخضر بقي شبه سائدا على معظم نماذج السايلج مع انبعاث رائحة الخل. كما تميزت النماذج بتماسك جيد فضلا عن وجود طفيف للتعفن في بعض النماذج التي لم تعامل باليوريا. واظهرت النتائج ايضا ان زيادة مستوى اضافة الحامض من 0.5 الى 1 و 1.5% ادت الى حصول زيادة ($P<0.01$) في المحتوى من المادة الجافة بلغت 0.79 و 1.15%, وفي البروتين الخام 1.42 و 2.11% على التوالي, وانخفاض ($P<0.01$) في مستخلص الايثر بنسبة 0.29 و 0.63%. اما تأثير مستوى اضافة اليوريا فان معظم المتغيرات قد اشارت الى حصول تراجع في التخمرات نظرا لحصول انخفاض معنوي في المادة الجافة والرماد ($P<0.01$) ومستخلص الايثر ($P<0.05$). كما انخفض المحتوى من الالياف الخام ($P<0.01$) الى 41.10% عند استخدام اليوريا بمستوى 2%, مقارنة مع 43 و 44.7% عند استخدام اليوريا بمستوى 1 و 0% على التوالي. واظهرت النتائج ايضا حصول انخفاض ($P<0.01$) في قيم الأس الهيدروجيني من 5.90 الى 4.99 و 4.88 وتركيز نتروجين الامونيا من 1.19 الى 0.75 و 0.66 % من النتروجين الكلي في نماذج السايلج, وتركيز الاحماض الدهنية الطيارة الكلية من 6.56 الى 4.61 و 4.14 مليمول/100 غم مادة جافة نتيجة لإضافة حامض الفورميك بمستوى 0.5 و 1 و 1.5% على التوالي. فيما ادت زيادة مستوى اليوريا الى حصول ارتفاع معنوي ($P<0.01$) في معايير التخمرات, 5.02 و 5.06 و 5.70 للاس الهيدروجيني, 0.67 و 0.98 و 0.95 % من النتروجين الكلي لتركيز نتروجين الأمونيا و3.70 و 5.53 و 6.07 مليمول/100 غم مادة جافة لتركيز الأحماض الدهنية الطيارة الكلية على التوالي.

كلمات مفتاحية: القصب, سايلج, حامض الفورميك, يوريا, تخمرات

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INTRODUCTION

Since arable land is limited, there will be high correlation between human and animal diets, hence searching for un-conventional plants or new source of green crops to feed animals should be hardly taken into account. Improve animal feeding depending on local available feed sources could enhance animal production via lowering production cost and minimizing deficiency in protein and energy. Common reed, *Phragmitis communis*, was the most expanded plant. Recently Al-Sultani (3) concluded that it was possible to produce good quality reed silage by addition of molasses and urea at level of 10 and 1% respectively. Silage is plant material exposed to fermentation in the silo. Ensiling is a conservation forage crops with minimum nutrient loss (1). It is based on natural fermentation under anaerobic condition in which epiphytic lactic acid bacteria (LAB) convert water soluble carbohydrates (WSC) into organic acid, as a result, pH decreases and the forage is preserved (30). To improve the ensiling process, various chemical and biological additives have been developed. Formic acid (FM) was known as effective additive. Addition of FA to silage material has been reported to have generally positive effects (42). The use of FA has been found to reduce pH, lactic acid, acetic acid and butyric acid in different kinds of silage (25, 26). Kennedy (16) reported that FA can restrict the fermentation by its ability to decrease pH and antibacterial effect that results in a limited fermentation and reduction in organic acid. The restriction of silage fermentation by FA is positively associated with higher content of residual WSC and lower proteolysis (15). Moreover, addition of FA effectively improved utilization of nutrients in silage (24). Improved silage preservation, *in vitro* digestibility and reduced in-silo losses (19). Molasses may be added with FA to take advantage on silage fermentation. Molasses enriches the fresh material with WSC and fills the gaseous pores, thereby reducing the influx of oxygen in the silage (7). The current study aimed to evaluate the effect of addition different levels of FA and urea as an available and cheap source of nitrogen (N) on nutritive value of wild reed silage through changes in chemical composition and fermentation nature.

MATERIALS AND METHODS

Making reed silage

This study was carried out in nutrition laboratory based on preparing 400-500 g samples of common reed silages with 4 replicates. Reed plants (40.84% DM, 15.22% ash, 5.71% CP, 3.74% EE, 44.95% CF and 30.38% nitrogen free extract (NFE)) were obtained from the area nearby Animal Production Department- Alqasim Green University. Plants were chopped into 2-3 cm of length. Treatment solutions were prepared by addition of date juice as a source of WSC at level of 10%, and three levels of both FA, 0.5, 1 or 1.5% and urea as a source of N, 0. 1 or 2%. Quantities of additives were estimated on DM basis of reed plants, tap water was added to ensure DM content of about 30% in treated materials. Samples were tightly packed in double nylon bags, compacted by hand to exclude the air. Samples were then moved to pit silos, well covered, filled up with soil and stored for 60 days. By the of this period bags of samples were moved again to laboratory to perform sensory characterization, chemical and fermentation analysis.

Sensory and fermentation characteristics

Sensory characteristics of silage included color, odor, aggregation and presence of molds were determined as described by Saeed (32). Fermentation characteristics included pH, NH_3-N and TVFA concentrations were determined in silage extract of each sample prepared as described by Levital et al. (18). pH was measured immediately using a pH meter. Concentrations of NH_3-N and TVFA were determined in preserved silage extract according to AOAC (4) and Markham (21), respectively.

Chemical analysis

Chemical analysis was performed in duplicate manner using methods of AOAC (4). DM content was determined by drying samples in oven at 105 ºC for 24 hours (h). Dried samples were left to cool in desiccator, grind and kept in plastic containers. Ash content was determined by ashing samples in furnace at 500 ºC for 4 h. Crude protein (CP) content was determined using S4 Kjeltec system manufactured by German Behr Company. EE content was determined by extraction with hexane in Soxhlet apparatus manufactured by

Korean FINE TECH, SCMP:F50-6H Company. CF content was determined using DOSI-Fiber Extractor manufactured by Spanish Selecta Company. NFE was determined by difference.

Statistical analysis

Data obtained were analyzed as a factorial experiment in completely randomized design by analysis of variance using statistical analysis system, SAS (38).

RESULTS AND DISCUSSION

Sensory characteristics

Table 1 shows sensory characteristics of reed silages as affected by levels of FA and urea. Most silage samples prepared by addition of FA at level of 0.5% without urea were greenish yellow, but addition of urea at 1 and 2% caused a light darkness, however, green color was still dominant. Similar dark green color was shown in samples prepared by addition of other levels of formic acid without urea. Other samples of reed silage showed colors ranged between greenish and yellowish. Similar observations were obtained by Al-Sultani (3) in which reed plants ensiled without or with 1% of urea were greenish yellow, whereas, those ensiled with higher level (2%) showed darker color. Degradation of urea during ensiling may affect color of silages in a current study. The slight differences in color may be due to dissociation of chlorophyll green color occurred during silage fermentation (10). Rowghani and Zamiri (30) reported that differences in concentration of organic acid as affected by addition of FA may interfere with the above observations.

Regarding odor, all samples characterized with diluted to concentrated vinegar odor. This may refer to existence of different levels of organic acids as evidenced by fermentation characteristics shown in table 4. Since all samples were ensiled with similar level of WSC source, vinegar odor (though it differed in strength) was affected by level of FA rather than level of urea. Djordjević et al. (12) indicated that increased level of FA increased acidity of silage. Those workers considered acidity as a result of dissociation of lactic and acetic acids produced during fermentation in addition to added FA. Odor observation was in line with that reported by Al-Sultani (3) in reed silage prepared without urea. Similar finding was also observed in most samples of reed silage in another study (32). Results revealed that most samples were well aggregated as a result of restriction of fermentation due to addition of FA. Slight presence of mold was observed in some samples especially those prepared without addition of urea. This can be explained by the antifungal role of ammonia released from dissociation of urea during ensiling (17). Similar result was obtained by Saeed and Al-Sultani (34), moldiness was observed in urea-untreated reed silage, whereas, 2% urea-treated reed silage was clear. Clearance of mold in reed silage in a current study may also correlated with level of FA, since it limits the fermentation by acidification. Antimicrobial effect of FA may interfere with cell function and inhibit growth of both mold and bacteria (20).

Table 1. Sensory characteristics of reed silages

Formic acid (%)	**0.5**			**1**			**1.5**		
Urea (%)	**0**	**1**	**2**	**0**	**1**	**2**	**0**	**1**	**2**
Color*	**GY**	**DG**	**DG**	**DG**	**GB**	**YB**	**DG**	**YB**	**YB**
Odor**	**DFV**	**DFV**	**DFV**	**DFV**	**CFV**	**DFV**	**DFV**	**DFV**	**DFV**
Aggregation***	**A**	**A**	**A**	**A**	**A**	**A**	**A**	**A**	**A**
Moldiness ******	+	–	–	+	–	–	+	–	–

*** GY: greenish yellow, DG: dark green, GB: greenish brown, YB: yellowish brown**

**** DFV: diluted fruit vinegar, CFV: concentrated fruit vinegar**

***** A: aggregated **** + , – : presence or absence of mold**

Chemical composition

Table 2 shows the effect of levels of FA and urea on chemical composition of reed silages. Statistical analysis revealed that except ash, chemical composition of reed silage was significantly affected by increasing level of FA. Increasing level of acid from 0.5 to 1 and 1.5% increased ($P<0.01$) DM content by 0.79 and 0.48% respectively. Increase DM content may be due to the restriction of fermentation by FA, hence samples prepared with higher levels of acid may recovered higher DM.

Similar results were obtained by many other studies (30,14, 16). Results showed significant ($P<0.01$) increase in CP content of reed silage with increasing level of FA, increases were 1.42 and 3.11% due to increasing level of acid from 0.5 to 1 and 1.5% respectively. Rowghani and Zamiri (30) reported similar results, and attributed higher CP content of FA treated whole corn crop silage to restricted effect of this acid on fermentation. Rooke et al. (29) clarified that this finding could be due to the restriction of fermentation, deamination and decarboxylation of proteins. Moreover, Selwet (40) illustrated that silages treated with organic acids were characterized with higher protein content probably due to limited growth of microorganisms leading to reduce intensity of protein proteolysis. Results of chemical composition showed as well that there was a significant ($P<0.01$) decrease in EE content of reed silages by 0.29 and 0.63% due to addition of FA at 1 and 1.5% as compared with 0.5% levels. This decrease may be due to restriction of fermentation caused by high levels of acid as indicated by fermentation characteristics (table 4). Worth mentioning that high silage content of fat may associated with existence of TVFA produced by anaerobic oxidation of WSC during ensiling (5). Regarding effect of urea levels on chemical composition of wild reed silage, results suggested that fermentation may be restricted. There was a significant ($P<0.01$) reduction in content of DM, 21.90, 21.12 and 20.70%; ash, 14.58, 12.18 and 11.53% and EE ($P<0.05$), 4.07, 3.92 and 3.77% in samples prepared without and with 1 and 2% of urea respectively. Reduction in EE content may be associated with DM loss occurred during ensiling. Savoie et al. (39) reported that DM loss through effluent during ensiling may be up to 16%. Al-Sultani (3) found that ensiling reed plants with urea at level of 2% caused DM loss of 2.48%, however, those ensiled with 1% of urea recovered higher DM. The inconsistency with a current study when urea was added at level of 1% may be attributed to quantity of water used at ensiling. Water in study of Al-Sultani (3) was added to reduce DM content of reed plants to about 40%, whereas, much water was required in a current study to reduce DM content to about 30%. Saeed and Mohammed (36) noticed that increase level of urea added at ensiling corn cobs caused a significant ($P<0.01$) reduction in EE content. Workers explained that by restriction of fermentation caused by higher ammonia concentration produced from dissociation of urea during storage leading to increase pH. Similar changes were observed in values of pH and NH_3-N in a current study (table 4). As expected there was ascending trend in CP content, higher ($P<0.01$) values were recorded when higher level of urea was added, this agreed with many studies (33, 41, 32). Existence of ammonia produced from dissociation of urea during ensiling may explained this increase (37). This was confirmed in a current study by significant ($P<0.01$) increase in NH_3-N concentration with increasing urea levels added at ensiling from 0

Table 2. Effect of levels of formic acid and urea on chemical composition of reed silages (% ± SE).

Item	Level of formic acid (%)			Level of urea (%)			P	
	0.5	1	1.5	0	1	2	FA	U
DM	20.59^{b} ± 0.23	21.38^{a} ± 0.33	21.74^{a} ± 0.26	21.90^{a} ± 0.31	21.12^{b} ± 0.25	20.70^{b} ± 0.26	**	**
% in DM								
Ash	12.83 ± 0.36	12.66 ± 0.45	12.80 ± 0.46	14.58^{a} ± 0.16	12.18^{b} ± 0.18	11.53^{c} ± 0.17	NS	**
CP	4.91^{c} ± 0.15	6.33^{b} ± 0.31	7.02^{a} ± 0.30	5.55^{b} ± 0.09	5.88^{b} ± 0.33	6.83^{a} ± 0.47	**	**
EE	4.23^{a} ± 0.10	3.94^{b} ± 0.06	3.60^{c} ± 0.07	4.07^{a} ± 0.10	3.92^{ab} ± 0.13	3.77^{b} ± 0.07	**	*
CF	43.09 ± 0.70	43.51 ± 1.04	41.97 ± 1.27	44.47^{a} ± 1.07	43.00^{ab} ± 0.85	41.10^{b} ± 0.96	NS	**
NFE	34.94 ± 0.76	33.56 ±1.53	34.61 ± 1.39	31.33^{b} ± 1.12	35.02^{a} ± 1.00	36.77^{a} ± 0.95	NS	**

Means with different letters at the same row are significantly different at * ($P<0.05$) ** ($P<0.01$)

to 1 and 2%, values were 0.67, 0.98 and 0.95% of total N respectively (table 4). Statistical analysis revealed that CF content was significantly decreased ($P<0.01$) when higher level of urea was added, values were 41.10 as compared with 43 and 44.7% in samples of reed silages prepared with lower level and without addition of urea respectively. Similar result was obtained by Saeed (32) in reed silage due to addition of urea. Saeed (33) attributed reduction in CF content of wheat straw silage to the partial degradation of carbohydrate complexes due to increased activity of silage microbes. He suggested that these microbes may be provided with suitable N sources (urea) in existence of encouraging, but insufficient source of fermentable carbohydrate (molasses), hence they compelled to degrade energy complexes of cell walls. Moreover, reduction in CF content can be explained by role of ammonia released from degradation of urea during ensiling. Celic et al. (11) reported that ammonia may help breaking down bonds linked cellulose and hemicellulose with lignin in cell wall skeleton. Results of a current study showed that NFE content was significantly increased ($P<0.01$) due to addition of urea. Lower ($P<0.01$) NFE content was found in samples of reed silages prepared without addition of urea (31.33), whereas, higher ($P<0.01$) content was found in samples prepared with addition of 1 and 2% levels of urea (35.02 and 36.77% respectively). These increases may be due to effect of ammonia produced during ensiling on degradation of cell components (32, 11). Table 3 shows effect of interaction between levels of FA and urea on chemical composition of reed silages. Significant analysis showed that DM and EE contents were affected ($P<0.05$) by this interaction in similar trend of main effect of both factors. It seemed that samples of reed plants ensiled with FA at level of 1% without (21.99%), or with addition of low level of urea (21.87%) recovered higher DM in comparison with other samples, especially, those prepared with addition of high level of urea. This may reflect the restricted effect of acid (8, 30, 27). This effect may associated with lower DM loss. Samples of reed silages prepared with addition of FA at low (0.5%) and mid (1%) levels had higher EE content (4.32- 4.57%) as compared with other samples, especially those prepared with high level of acid (3.51-3.71%). This may reflects the role of FA on silage fermentation, in which higher levels were shown to be more effective. Considering that WSC was converted into fatty acids during aerobic fermentation (5). Fat content may be expected to decrease due to reduction of VFA produced as a result of restriction of fermentation as shown in table 4. Statistical analysis of interaction effect also showed higher ($P<0.01$) CP content in reed silages prepared with addition of urea at low and high levels and FA at high (7.21 and 8.13% respectively) and medium (7.63%) levels. This result was consistent with the synergized effects of: 1) Urea which increased N and CP contents of silages with each increase in its level (33, 41, 32). 2) Formic acid which may limited fermentation rate and consequently, reduced protein degradation (40). Results revealed lower ($P<0.01$) CF content in samples of reed silages prepared with higher level of FA and urea (37.65%). This lower content though it was not significantly differed with many other samples, it can be explained by the effect of ammonia released from dissociation of urea during 60 days-ensiling period (11, 32), together with the effect of FA (table 2).

Characteristics of silage fermentation

Table 4 shows effect of levels of FA and urea on fermentation characteristics of reed silages. Statistical analysis revealed that pH was significantly ($P<0.01$) decreased with increasing level of formic acid. Similar result was obtained by Hapsari et al. (13) in grass silage. Reduction of pH can be explained on basis of the strength of formic acid which is twice stronger than lactic acid (3.75 vs. 3.85 pKa) (43). Lorenzo and O'Kiely (19) reported that FA lowered the pH immediately after its addition and worked by reducing the activity of saccharolytic enterobacteri and clostridia bacteria. Baytok and Muruz (7) concluded that addition of FA at level of 0.5% with 2, 4 and 6% levels of molasses produced well preserved grass silages with low pH, 4.62, 4.51and 4.54 respectively.

Table 3. Effect of interaction between levels of formic acid and urea on chemical composition of reed silages (% ± SE).

Level of formic acid (%)	0.5			1			1.5			P	SE
Level of urea (%)	0	1	2	0	1	2	0	1	2		
DM	21.10^{ab}	20.62^{b}	20.06^{b}	21.99^{a}	21.87^{a}	20.29^{b}	21.98^{a}	20.17^{b}	21.06^{ab}	*	± 0.16
% in DM											
Ash	14.46^{a}	9.98^{d}	12.04^{b}	14.69^{a}	9.70^{d}	11.60^{bc}	14.58^{a}	10.86^{c}	10.96^{c}	**	± 0.32
CP	5.44^{c}	4.57^{d}	4.72^{d}	5.48^{c}	5.86^{c}	7.63^{ab}	5.73^{c}	7.21^{b}	8.13^{a}	**	± 0.21
EE	4.43^{a}	4.45^{a}	3.81^{b}	4.56^{a}	4.32^{a}	4.43^{a}	3.71^{b}	3.51^{b}	3.59^{b}	*	± 0.17
CF	41.68^{bc}	43.23^{ab}	44.37^{ab}	47.56^{a}	41.69^{bc}	41.29^{bc}	44.18^{ab}	44.09^{ab}	37.65^{c}	**	± 0.59
NFE	33.99^{bc}	37.77^{ab}	35.06^{abc}	27.71^{d}	38.43^{ab}	35.05^{abc}	31.80^{cd}	34.33^{bc}	39.67^{a}	**	± 0.72

Means with different letters at the same row are significantly different at * (P<0.05) ** (P<0.01)

In a current study, increasing level of FA from 0.5 to 1 and 1.5% decreased (P<0.01) pH of reed silages from 5.90 to 4.99 and 4.88 respectively. The decrease in pH values associated with the increase of FA level was attributed to the dissociation of the produced lactic and acetic acid and also of the FA used (12). Results revealed that there was a significant (P<0.01) decrease in NH_3-N concentration in reed silage with increasing level of FA. Values were 1.19, 0.75 and 0.66% of TN in samples prepared with addition of 0.5, 1 and 1.5% levels of FA. This may be attributed to restriction of fermentation as affected by the limited effect of acid (8, 30, 27). Since low pH limits process of protein degradation during ensiling by inhibiting plant proteolytic enzymes (22). Then, reduction in pH of reed silages due to addition of FA may be another reason for reduction of NH_3-N concentration observed in a current study. Concentrations of TVFA were changed with similar trend as NH_3-N concentrations. Values were significantly (P<0.01) decreased from 6.56 to 4.61 and 4.14 mmol/100 g DM with increasing levels of FA from 0.5 to 1 and 1.5%, respectively. This result confirmed that addition of acid limited silage fermentation. Similar result was obtained by Saarisalo et al. (31), in this study addition of FA resulted in immediate reduction in concentrations of NH_3-N and TVFA in grass silage. Workers attributed reduction in these fermentation parameters to the restriction of silage fermentation caused by addition of FA.

Table 4. Effect of levels of formic acid and urea on fermentation characteristics of wild reed silages (% ± SE).

Item	Level of formic acid (%)			Level of urea (%)			P	
	0.5	1	1.5	0	1	2	A	U
pH	5.90^a ± 0.27	4.99^b ± 0.12	4.88^c ± 0.04	5.02^b ± 0.13	5.06^b ± 0.16	5.70^b ± 0.29	**	**
NH_3-N % of TN	1.19^a ± 0.14	0.75^b ± 0.02	0.66^b ± 0.02	0.67^b ± 0.02	0.98^a ± 0.12	0.95^a ± 0.12	**	**
TVFA mmol/100 ml	6.56^a ± 0.76	4.61^b ± 0.24	4.14^b ± 0.36	3.70^b ± 0.14	5.53^a ± 0.23	6.07^a ± 0.85	**	**

Means with different letters at the same row are significantly different at * (P<0.05) ** (P<0.01)

Regarding effect of levels of urea, results showed that there was a significant (P<0.01) increase in pH of reed silage with increasing level of urea. Values were, 5.02, 5.06 and 5.07 in samples of silages prepared without and with addition of urea at levels of 1 and 2% respectively. The increase in pH of silage with increasing urea levels can be explained on basis of basic effect of ammonia released from the rapid dissociation of urea during ensiling (37, 35). Ammonia concentration certainly increased as level of urea increased. This result agreed with that reported by Saeed (32) in which higher (P<0.05) pH was recorded in samples of reed silage from 4.56 to 4.93 due to addition of urea at level of 3%. Addition of FA at levels of 0.5, 1 and 1.5% in a current study may explained the increase in pH values with respect to the previous study. Because of indirect role of FA in limiting silage fermentation (31). Through reduction in concentration of lactic acid (13). However, Aksu et al. (2) demonstrated that lactic acid concentration was not affected by addition of FA. Moreover, it was suggested that FA applied at moderate rates is particularly effective at inhibiting the activity of undesirable bacteria, thereby, it may permit lactic acid bacteria to dominate the fermentation (22, 28). Saeed and Al-Sultani (34) reported similar trend to the increase in pH of silage observed in a current study, however, lower values of pH were obtained in that study, 3.75, 4.30 and 6.70 in reed silage prepared without and with addition of 1 and 2% of urea. Difference in circumstances of ensiling may explained the differences in pH values, in a current study samples of reed sila--ge were prepared in laboratory using hands to compact treated materials, whereas, in the other study, silages were made in a field using a tractor to insure compaction. As expected, statistical analysis of fermentation data showed that there was a significant (P<0.01) increase in NH_3-N concentrations in samples of reed silages with increasing levels of urea. Values were 0.67, 0.98 and 0.96 % of TN. This increase may be attributed to the probable increase in dissociation of urea as a result of microbial urease activity (37). Concentrations of TVFA were significantly ($P<0.01$) increased by 2.42 and 1.87 mmol/100 g DM in samples of reed silages prepared with 1 and 2% levels of urea as compared with those prepared without addition of urea. Azim et al. (6) indicated that urea-treated oat silage characterized with high concentration of TVFA. Muck (23) reported that about 60% of studies he surveyed mentioned to the increase in fermentation acids due to addition of non-protein nitrogen (NPN) sources at ensiling. Moreover, Bolsen et al. (9) found that an increase in concentration of lactic acid was detected as a result of addition 0.5% of urea or 0.35-0.4% of ammonia. Effect of interaction between levels of FA and urea on fermentation characteristics of reed silages was shown in table 5. Lower pH values were recorded in samples of reed silage prepared with higher level of FA regardless to levels of urea, 0, 1 or 2% (4.80, 4.87 and 5.98, respectively), samples prepared with lower level of acid without urea (4.88), and those prepared with medium level of acid and lower level of urea (4.88). Samples prepared with lower level of acid and urea at levels of 1 and 2% were characterized with higher concentrations of NH_3-N, 1.52 and 1.51 % of TN, respectively. This may be attributed to dissociation of urea in medium of lower level of FA, in which degradation of protein may not be effectively inhibited. Higher concentration of TVFA was recorded in samples prepared with addition of 0.5% level of FA and 2% level of urea (9.82 mmol/100 g DM), whereas, lower concentrations were recorded in samples of reed silages prepared without addition of urea and all levels of FA. This can be explained on basis of integration of lesser effect of lower level of acid as compared with other levels in restricting fermentation and consequently slight reduction in acid produced during fermentation (31), and effect of addition of high level of urea which enhanced acid production.

Table 5. Effect of interaction between levels of formic acid and urea on fermentation characteristics of reed silages (as appeared ± SE).

Level of formic acid (%)	0.5			1			1.5			P	SE
Level of urea (%)	0	1	2	0	1	2	0	1	2		
pH	4.88^{de}	5.77^{b}	7.07^{a}	5.39^{c}	4.55^{e}	5.05^{d}	4.80^{de}	4.87^{de}	4.98^{d}	**	± 0.12
NH_3-N, mg/100 ml	0.55^{c}	1.52^{a}	1.51^{a}	0.72^{bc}	0.75^{b}	0.80^{b}	0.75^{b}	0.67^{bc}	0.55^{c}	**	± 0.06
TVFA mmol/100 ml	3.91^{cd}	5.96^{b}	9.82^{a}	3.88^{cd}	5.07^{bc}	4.86^{bc}	3.32^{d}	5.57^{b}	3.54^{d}	**	± 0.33

Means having different letters at the same row are significantly different at * ($P<0.05$) ** ($P<0.01$)

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