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# Oxalate Accumulation in Forage Plants: Some Agronomic, Climatic and Genetic Aspects

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**ABSTRACT :** Oxalic acid is synthesized by a wide range of plants. A few of them are forage plants that can cause oxalate poisoning in ruminants under certain conditions. In this paper, the role of some agronomic, climatic and genetic factors in minimizing oxalate accumulation in forage plants has been discussed. Research indicates that the content of oxalate in forage can be controlled by fertilizer application. For example, nitrate application resulted in higher contents of soluble and insoluble oxalates than ammonium application. With an increased rate of potassium application, soluble oxalate content showed an increasing trend and insoluble oxalate content showed a decreasing trend. With an increased rate of calcium application, soluble oxalate content showed a decreasing trend and insoluble oxalate content showed a reverse trend. Other agronomic factors such as growing season, harvesting practices, plant maturity, plant species, plant variety and plant parts can also have a large effect on oxalate accumulation. However, the potential benefits of the above approaches for improving forage quality have not been fully exploited. In addition, there is still insufficient information to fully utilize means (e.g. plant nutrients, season and soil moisture) to minimize oxalate accumulation in forage plants. Therefore, more research is required for a better understanding of the interactions between oxalate and the above-mentioned factors in forage plants. (**Key Words :** Agronomic Practices, Fertilization, Forage, Oxalate Accumulation)

# INTRODUCTION

Many forage plants such as setaria (Setaria sphacelata), buffelgrass (Cenchrus ciliaris), pangolagrass (Digitaria decumbens), kikuyugrass (Pennisetum clandestinum) and napiergrass (Pennisetum purpureum) represent a major food source for livestock in the tropics and subtropics. However, these species can sometimes accumulate oxalate (as soluble and/or insoluble form) to potentially toxic concentrations (Dhillon et al., 1971; Cheeke, 1995; Rahman et al., 2006). In general, oxalate poisoning is a complex issue. Factors such as chemical form of the oxalate, age of the animal, adaptation of animals to oxalate-rich forage, composition of the diet and availability of water for animals could influence the susceptibility of animals to oxalate poisoning. Ingested oxalate complexes with dietary calcium (Ca) and forms insoluble Ca oxalate. This leads to disturbances in Ca and phosphorus (P) metabolism involving excessive mobilization of bone mineral. The demineralized bones become fibrotic and misshapen, causing lameness and "bighead" in horses (McKenzie et al., 1981). Ruminants are less affected, but prolonged grazing by cattle and sheep on some tropical grass species can result in severe hypocalcaemia (Seawright et al., 1970).

When a ruminant consumes an oxalate-containing plant, the oxalate is metabolized in four possible ways. First, soluble oxalate may be degraded by rumen microflora (Allison et al., 1977). Second, soluble oxalate may combine chemically with Ca to become insoluble oxalate, which is excreted in feces, reducing absorption of Ca. Third, soluble oxalate may be absorbed from the rumen into the blood stream where it can combine with serum Ca to form insoluble oxalate crystals (Blaney et al., 1982). This crystal may then precipitate in the kidneys and can cause kidney failure (Lincoln and Black, 1980). Fourth, ingested insoluble oxalate from plants may pass through the digestive tract without a harmful effect on the body's metabolism (Ward et al., 1979).

Oxalate is a common constituent of plants. Studies have shown that oxalate may play various roles in plants, including Ca regulation, ion balance, plant protection, tissue support and heavy metal detoxification (Libert and Franceschi, 1987). Soluble oxalate usually forms with sodium (Na<sup>+</sup>), potassium (K<sup>+</sup>) and ammonium (NH<sub>4</sub><sup>+</sup>) ions,

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and insoluble oxalate forms with  $Ca^{2+}$ , magnesium (Mg<sup>2+</sup>) and iron (Fe<sup>2+</sup>) ions (Savage et al., 2000). Soluble oxalate content in napiergrass was correlated with K concentration, while insoluble oxalate content was correlated with Ca and Mg concentrations (Rahman et al., 2008b).

Several pathways for oxalate production have been proposed, including photorespiratory glycolate/glyoxylate oxidation, cleavage of ascorbate and hydrolysis of oxaloacetate (Nakata, 2003). An oxalate biosynthetic pathway in plants remains controversial, but more researchers tend to the view that the predominant biosynthetic pathway of oxalate may vary from one plant species to another. A review on oxalate in crop plants has been summarized by Libert and Franceschi (1987). Studies on some agronomic, climatic and genetic approaches affecting oxalate content in forage plants have been carried out from time to time, but it is difficult to compare the results of one study with another due to sporadic research. The present review aims to summarize those factors which alter the oxalate content in forage plants.

# FERTILIZER/ELEMENT APPLICATION

In general, 16 nutrients are essential for plant growth and development. Of the major nutrients, nitrogen (N), P and K are required in relatively large amounts. The nutrients Ca, Mg and sulfur are also required in relatively large amounts, but are less likely to be deficient in the soil system. Micro-nutrients (e.g. iron, chlorine, manganese, boron etc.) are essential to plants in relatively small amounts. Some of these nutrients, including N, K, Na and Ca are partially involved in oxalate accumulation in forage.

#### Nitrogen fertilization

Several types of N fertilizer are available, including urea, ammonium nitrate, ammonium sulfate and manure. Application of urea to forage did not result in excessive oxalate accumulation (Williams et al., 1991; Rahman et al., 2008a, 2008b; Rahman et al., 2009b). However, these observations contradict the result of Jones and Ford (1972) who reported that the oxalate content in setaria increased from 3.3% to 5.6% as N fertilizer (as urea) level increased from 0 to 200 kg/ha, respectively. Roughan and Warrington (1976) reported that setaria accumulated a similar content of oxalate irrespective of whether ammonium, urea or nitrate was the sole source of N. These results suggest that setaria is a grass in which oxalate accumulation responds markedly to application of any form of N.

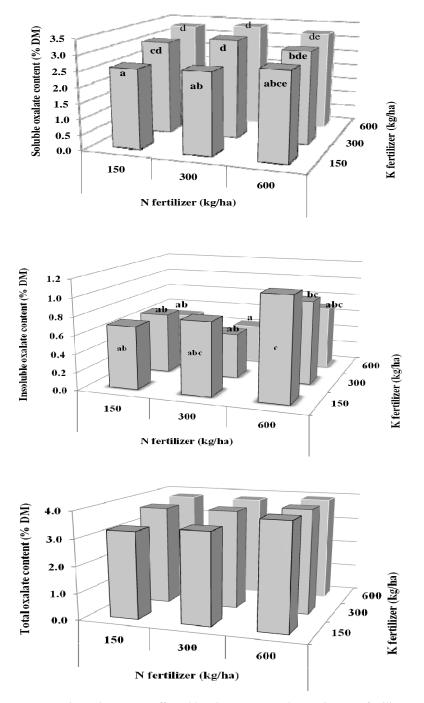
Kipnis and Dabush (1988) showed that oxalate content in napiergrass was not affected by application of N as ammonium nitrate. Similar results were found by Rahman (2009). Information on the effects of ammonium sulfate or manure on oxalate content is still lacking.

Nitrate application resulted in higher oxalate accumulation in rice and some vegetable crops than did ammonium application (Ji and Peng, 2005; Zhang et al., 2005; Xu et al., 2006). Recently, a nutrient solution study suggested that similar effects of nitrate-N are also observed in forage plants (Rahman et al., 2010b). It is thought that when N is provided as nitrate in plants, a number of hydroxyl (OH<sup>-</sup>) ions are produced by catalysis with nitrate reductase and nitrite reductase. The increased levels of OHions may act as a signal to stimulate organic acid biosynthesis (e.g. oxalic acid). As a result, the H<sup>+</sup> ion dissociated from the organic acid can neutralize the excess levels of OH<sup>-</sup> ion in order to maintain intracellular pH (Raven and Smith, 1976; Allen et al., 1988), and the remaining organic (COO<sup>-</sup>) ions may stimulate cation uptake (e.g. Na, Ca and Mg). On the other hand, when plants are grown with ammonium, the cytoplasm acidifies due to excretion of H<sup>+</sup> ion (Schubert and Yan, 1997), which probably inhibits organic acid biosynthesis.

Both urea and ammonium nitrate contain ammonium-N, and while in plants ammonium may serve as a negative signal inhibiting oxalate accumulation, the studies above indicate that fertilization with urea and ammonium nitrate may not influence the oxalate content in forage. Further studies are necessary to determine the precise role of nitrate and nitrate reduction-related enzymes (e.g. nitrate reductase, glyoxylate dehydrogenase or glycolate dehydrogenase) in regulation of oxalate in plants through physiological, biochemical and molecular approaches.

### **Potassium fertilization**

Concentrated animal operations generate substantial amounts of manure that is rich in plant nutrients, including K. Excessive manure is often applied on limited areas of forage crop fields resulting in high or excessively high levels of K (Sunaga et al., 2005). Experiments carried out with napiergrass showed that soluble oxalate content correlates highly with K concentration (Rahman et al., 2008b). Smith (1972) reported that total oxalate content in setaria increased with an increased rate of application of K. In another study, Smith (1978) observed that oxalate content tended to increase with applied N, but exhibited a strong N×K interaction, declining with applied K when 57 mg N/pot was applied, remaining constant with 141 mg N/pot, and increasing with applied K at 283 mg N/pot. Rahman et al. (2010a) reported that soluble oxalate content in napiergrass appeared to increase while insoluble oxalate content appeared to decrease with an increased rate of K application, and significant interactions between N and K fertilizations were observed (Figure 1). In contrast, application of KCl to kikuyugrass did not affect the soluble oxalate content (Williams et al., 1991), which may be explained by chloride anions competing for cations and



**Figure 1.** Oxalate content (% DM) in napiergrass as affected by nitrogen (N) and potassium (K) fertilizer application (kg/ha). Values are expressed as average of 3 cuttings (Rahman et al., 2010a). The letters at the tops of the bars indicate statistical significance; means with different letters are significantly different (p<0.05).

depressing oxalate synthesis. Overall, application of K fertilizer with moderate or high levels of N enhances oxalate accumulation in forage. Therefore, K fertilization should be kept at a low level to avoid excess levels of soluble oxalate.

## **Phosphorus fertilization**

Information on the effect of P fertilization on oxalate

content in forage plants is still lacking. Phosphorus fertilizer as superphosphate depressed oxalate levels in vegetable plants (Singh, 1974). Adebooye and Oloyede (2007) showed that the oxalate content in *Trichosanthes cucumerina* was not affected by P levels. If plants are to grow in P-limited soils, they need mechanisms to access soil P. One proposed mechanism for P uptake is the production of oxalic acid by plants. For example,

Type of oxalate	Concentrations of NaCl (mM)				
	0	100	300	600	900
Soluble	1.15	1.35	1.22	1.16	1.15
Insoluble	0.31 <sup>ab</sup>	0.44 <sup>b</sup>	0.35 <sup>ab</sup>	0.37 <sup>ab</sup>	0.15 <sup>a</sup>
Total	$1.46^{ab}$	1.79 <sup>b</sup>	1.57 <sup>ab</sup>	1.53 <sup>ab</sup>	1.30 <sup>a</sup>

Table 1. Effect of salinity on oxalate content (% DM) in napiergrass (Rahman et al., 2008c)

<sup>a, b, c</sup> Means in the same row with different superscripts differ (p<0.05).

solubilization of soil P was demonstrated after oxalate addition in two soil types (Fox and Comerford, 1992). Cannon et al. (1995) observed that the total P concentration of the soil decreased with increasing oxalate. The above results show that oxalate may have an important link in P cycling, but that application of P may not increase the oxalate content in plants.

#### Salinity

There are a range of plants that grow in saline soils and which have been used as animal feed. It is not unusual for plants growing in saline areas to accumulate secondary compounds (e.g. oxalate, tannins etc.), and these can adversely affect palatability, feed intake and animal health (Masters et al., 2001; Masters et al., 2005). Halophytic plants such as saltbush (Atriplex halimus and Atriplex nummularia) contain high levels of secondary metabolites, mainly oxalic acid (Alazzeh and Abu-Zanat, 2004). The soluble oxalate content in halogeton (Halogeton glomeratus) was greatly increased by adding NaCl to the nutrient solution, and it was shown that Na is more effectively used in the accumulation of soluble oxalate than K (Williams, 1960). However, some studies failed to produce higher oxalate accumulation at higher levels of salinity (Williams et al., 1991; Rahman et al., 2008c). Rahman et al. (2008c) observed that 100 mM NaCl application produced slightly higher soluble oxalate content than in plants without additional NaCl application, but this value tended to decrease with further (from 300 to 900 mM NaCl) increases in NaCl application (Table 1). This result may be explained by a salinity effect on physiological responses of plants. For example, Gul et al. (2000) reported that net photosynthesis increased at low salinity (200 mM NaCl), but photosynthesis at higher salinity levels (400, 600, 800 and 1,000 mM NaCl) was not significantly different from the control (without NaCl). Fujii et al. (1993)

suggested that inhibition of photosynthesis may reduce the rate of biosynthesis of oxalate. In another study, Singh (1974) reported that if chloride or other anions are absorbed by plants, these anions compete for cations and depress oxalate synthesis. It seems that accumulation of oxalate in plants in the presence of salinity might be influenced by the chloride anion or photosynthesis rate.

### **Calcium fertilization**

Calcium fertilizers are occasionally applied to pastures, but little is known about the effect of Ca on oxalate content in forage plants. Recently, Rahman et al. (2009a) reported that soluble oxalate content in napiergrass showed a decreasing trend and insoluble oxalate content showed an increasing trend with an increased rate of Ca application (Table 2). Zindler-Frank et al. (2001) observed that soluble oxalate content in *Phaseolus vulgaris* plants decreased and insoluble oxalate content increased with increasing Ca concentration in the nutrient solution. Zindler-Frank et al. (2001) also observed that oxalate biosynthesis was not influenced by Ca. The above results suggest that the soluble oxalate content can be partially replaced by insoluble oxalate due to the use of Ca fertilizer.

# **CLIMATIC FACTORS**

### Season/temperature

Seasonal variation strongly affects the level of oxalate in saltbush plants. Abu-Zanat et al. (2003) observed that oxalate levels of *Atriplex* species were 8.29 and 4.92% of dry weight in spring and fall seasons, respectively. Plant tissue obtained in the early summer exhibited higher oxalate content when compared to similar samples obtained later in the season (Rahman et al., 2006). Rahman et al. (2006) also reported that leaf and stem tissues obtained in the early summer exhibited nearly equivalent levels of oxalate. As

Table 2. Effect of Ca(OH)2 on oxalate content (% DM) in napiergrass (average value of 3 cuttings; Rahman et al., 2009a)

Type of evaluate		Rate of Ca(C	$(g/m^2)$	
Type of oxalate	0	30	60	90
Soluble	1.15 <sup>b</sup>	1.09 <sup>b</sup>	$0.99^{ab}$	0.79 <sup>a</sup>
Insoluble	0.31 <sup>a</sup>	$0.46^{a}$	$0.58^{ab}$	$0.78^{b}$
Total	1.47	1.55	1.57	1.57

<sup>a, b</sup> Means in the same row with different superscripts differ (p<0.05).

the season advanced, the oxalate content in leaf tissue decreased gradually, whereas the decrease was more rapid in the stem tissue. Singh (2002) reported that the high values of oxalate observed in a napier-bajra hybrid during the month of June and July might be due to the peak in growth during summer and rainy seasons.

Tropical forages generally contain more oxalate than species grown in temperate regions, which implies a relationship between temperature and oxalate accumulation in plants. However, in a nutrient solution study, temperatures did not have any effect on the contents of total oxalate or soluble oxalate in napiergrass (Kipnis and Dabush, 1988). Many environmental effects associated with seasonal change (e.g. precipitation, day length, hours of sunlight, floral induction etc.) might alter oxalate levels. Based on the above results, it is still not known how season or temperature affects oxalate content in forage. Extensive research is required to understand the role of season or temperature on oxalate accumulation in forage plants.

# Soil moisture

Soil moisture can alter the level of oxalate in the plant. For example, under high soil moisture, *Atriplex halimus* plants were reported by Ellern et al. (1974) to accumulate higher levels of oxalate in leaves and twigs. Their data suggested that moisture stress sharply reduces insoluble oxalate and also may reduce oxalate levels during the fall season. Ala et al. (1995) reported that exceeding soil waterholding capacity (waterlogging) affected the oxalate content in *Atriplex amnicola* after 4 weeks of growth, with non-significant effects after 8 weeks of growth.

In contrast, the oxalate content of napiergrass was not related to soil moisture (Kipnis and Dabush, 1988). Soluble oxalate content in pot-grown napiergrass under unprotected cultivation was somewhat lower than the oxalate content under protected (inside a vinyl house) cultivation, especially in plant material sampled during the early summer (Rahman, 2009). This may have been due to excess soil moisture or nutrient run-off due to abundant early summer precipitation under unprotected cultivation. In contrast, soil conditions in pots under protected cultivation remained drier with a lower likelihood of nutrient run-off. It was shown that soluble oxalate content in pot-grown napiergrass inside a vinyl house was similar to results from field grown napiergrass (Rahman et al., 2006). As described above, the effect of soil moisture on oxalate content in forage plants is far from clear and more research is needed in this regard.

# **Diurnal variation**

Jones and Ford (1972) showed that samples taken during the day and night showed a diel pattern with lowest oxalate levels at midday and highest at night. In contrast, Roughan and Warrington (1976) failed to find diurnal variation in setaria. Libert and Franceschi (1987) suggested that photorespiration is not a prerequisite for oxalate synthesis. There are two known enzymes which can catabolize oxalate, namely oxalate oxidase and oxalate decarboxylase. Srivastava and Krishnan (1962) found a distinctive diel variation in the oxalate oxidase activity and oxalate content in the leaves of *Bougainvillea*. The enzyme activity of the leaves was at a minimum at 4 pm and at a maximum at 12 pm, whereas the total oxalate content was opposite that of the enzyme activity. Overall, diel variation in oxalate content in plants remains a subject of intense study. The diel variability in physiological processes is one of the endogenous rhythms operating in plants.

# **GENETIC FACTORS**

#### **Plant species**

Oxalate is found in almost all plant families. Research on oxalate content in various plants has shown significant genotypic variation (Table 3). Plants in the genera Halogeton, Rumex and Oxalis show high levels of oxalate accumulation. Plants in the genera Setaria, Cenchrus and Pennisetum show intermediate oxalate accumulation, whereas most other plant species show low levels of oxalate accumulation. The majority of plant species commonly consumed by livestock that accumulate oxalate contain much lower contents, typically around 1.0 to 2.0%. For example, Cenchrus ciliaris (buffalo hay) was reported to contain 1.1% soluble oxalate while the reported contents in rice straw (Oryza sativa) were 1.0 to 2.5% of dry weight (Libert and Franceschi, 1987). In another study, rice leaves contained 3.0-6.0% oxalate depending on growth stage and culture conditions (Ji and Peng, 2005); with 40-50% of the oxalate found in the soluble form (Xu et al., 2006). In many developing countries, rice straw is fed as a basal diet to ruminants.

There are many oxalate-containing plants that may cause poisoning to ruminants. They include setaria, halogeton, soursob (Oxalis spp.), pokeweed (Phytolacca americana), purslane (Portulaca oleracea), lamb's quarter (Chenopodium album), bassia (Bassia hyssopifolia), greasewood (Sarcobatus vermiculatus), pigweed (Amaranthus spp.), Russian thistle (Salsola kali), sugar beets (Beta vulgaris), rhubarb (Rheum rhaponticum) and various others (Jones et al., 1970; Cheeke and Shull, 1985). Halogeton contains 17 to 30% soluble oxalate per dry plant weight (Cronin and Williams, 1965). The leaves of fodder beet may contain up to 10% soluble oxalate (Libert and Franceschi, 1987). The importance of plant species that accumulate oxalate is clear from the literature. Plant species having a lower tendency to accumulate oxalate might be

Common name	Scientific name	Soluble oxalate	References
Halogeton	Halogeton glomeratus	12.8	James et al. (1976)
		17.0-30.0	Cronin and Williams (1965)
Setaria	Setaria sphacelata	1.2-6.3	Jones and Ford (1972)
		5.4-6.9	Jones et al. (1970)
Kikuyugrass	Pennisetum clandestinum	0.4-2.4	Marais (1997)
		1.8-2.2	Peet et al. (1990)
Kochia	Kochia scoparia	6.0-9.0	Undersander et al. (1990)
Buffelgrass	Cenchrus ciliaris	1.2-2.2	Playne (1976)
		3.5-4.3	Silcock and Smith (1983)
Napiergrass	Pennisetum purpureum	2.6	Garcia-Rivera and Morris (1955)
		3.0	Dhillon et al. (1971)
		3.3-3.6	Lal et al. (1966)
		1.8-3.8	Rahman et al. (2006)
Pearl millet	Pennisetum typhoides	1.3-3.0	Lal et al. (1966)
Guineagrass	Panicum maximum	1.1-2.3	Garcia-Rivera and Morris (1955)
Bassia	Bassia hyssopifolia	6.1	James et al. (1976)
Pigweed	Amaranthus retroflexus	4.5-9.4	Everist (1974)
Curly dock	Rumex crispus	6.6-11.1	Panciera et al. (1990)

Table 3. Soluble oxalate content (% DM) in some plant species

selected for cultivation; otherwise, consumption of oxalatecontaining plants by ruminants should be carefully monitored.

### **Plant variety**

Oxalate content varies among varieties within the same species. For example, of two cultivars (Nandi and Kazungula) of setaria, Nandi contains low oxalate, while Kazungula contains high oxalate (Jones and Ford, 1972). Abu-Zanat et al. (2003) observed that *Atriplex halimus* contained higher levels of oxalate (7.01%) compared with *Atriplex nummularia* (6.20%). Libert and Creed (1985) tested 78 rhubarb cultivars under the same growing practices and found oxalate ranged from 3.35% to 9.48% of dry weight; a threefold difference. Libert (1987) calculated that 72% of the variability in oxalate originated in genotype. Cultivars with high proportion of leaves may also have a higher oxalate level than varieties with low proportion of

leaves, since leaves normally contain higher oxalate than stems. For example, compared to a non-dwarf variety, a dwarf variety of napiergrass exhibited higher oxalate content, though the difference was not significant (p>0.05) (Rahman et al., 2006). These results show that oxalate content differs among cultivars and attention should be made to select those cultivars that accumulate a lower content of oxalate under the same growing conditions. Therefore, breeding of commercially viable low oxalate cultivars seems an approach to reducing oxalate in plants.

# **Plant parts**

The distribution of oxalate in plants is uneven (Table 4). Several researchers reported that oxalate content is highest in leaf tissue, followed by stem tissue (Jones and Ford, 1972; Marais et al., 1997; Rahman et al., 2006). Bamboo has three times the content of oxalate in younger parts of the shoot compared with older parts (Kozuke et al., 1983).

Species	Plant part	Soluble oxalate	References	
Setaria	Leaf blade	4.40	Jones and Ford (1972)	
	Leaf sheath	2.90		
	Stem	2.80		
Kikuyugrass	Leaf	1.33	Marais (1990)	
	Stem	0.39		
	Leaf	2.44	Marais et al. (1997)	
	Stem	0.98		
Napiergrass	Leaf	2.78	Rahman et al. (2006)	
	Stem	2.05		

Table 4. Soluble oxalate content (% DM) in plant parts of some forage species

In contrast, leaf and stem tissues exhibited nearly equivalent levels of oxalate when napiergrass was harvested in early summer (Rahman et al., 2006), possibly due to peak growth in summer and rainy seasons. Dhillon et al. (1971) reported that oxalate content in napiergrass (cv. Pusa Giant) is directly related to the thickness of the stem: the thicker the stem the higher the oxalate content.

Ruminants, particularly small ones, tend to prefer leaves rather than stems, but because leaves usually contain more soluble oxalate than the other parts of the plant, careful attention should be paid to plant materials consumed by grazing animals.

# **OTHER AGRONOMIC FACTORS**

### Maturity/harvesting practices

Young plants contain more oxalate than older plants (Jones and Ford, 1972). During early stages of growth, there is a rapid rise in oxalate content followed by a decline in oxalate levels as the plant matures (Davis, 1981). Rahman et al. (2009b) observed that the oxalate content of napiergrass can be manipulated by varying the harvesting interval, and that oxalate content declined as the harvest interval increased (Table 5). In contrast, in some plant species such as rhubarb (Rheum rhabarbarum), halogeton and water hyacinth (Eichhornia crassipes), oxalate content tends to increase as the plant matures. Halogeton becomes more toxic as growth proceeds, reaching a peak of toxicity at maturity (Torell et al., 2005). Abu-Zanat et al. (2003) observed that clipping of saltbush plants had no effect on their oxalate content, which was 4.99% in unclipped twigs versus 4.80% in regrowth. Abu-Zanat et al. (2003) also observed that seedlings of Atriplex nummularia contained more oxalate than old plants (9.65% and 7.63%), whereas old shrubs of Atriplex halimus (10.69%) contained more oxalate than the young seedlings (8.8%). Therefore, it is desirable to find harvesting practices in some forage plants that would reduce the oxalate content to acceptable levels by choice of appropriate growth stages for harvest.

# **Postharvest processing**

The effect of boiling or soaking on oxalate content of human food has been studied extensively. A marked decrease in oxalic acid content was reported in a starch

**Table 5.** Effect of clipping interval on oxalate content (% DM) innapiergrass (Rahman et al., 2009b)

Type of oxalate		Clipping interva	1
Type of Oxalate	2 weeks	4 weeks	8 weeks
Soluble	1.82 <sup>b</sup>	1.10 <sup>a</sup>	0.96 <sup>a</sup>
Insoluble	1.23	1.27	0.94
Total	3.04 <sup>c</sup>	2.38 <sup>b</sup>	1.89 <sup>a</sup>

<sup>a, b, c</sup> Means in the same row with different superscripts differ (p<0.05).

tuber (*Icacinia manni*) upon fermentation (Antai and Obong, 1992). Furthermore, Ologhobo (1989) observed that the oxalate content of soybean (*Glycine max*) seed was reduced by soaking and germination. Boiling can also reduce the soluble oxalate content of human food, if the cooking water is discarded (Noonan and Savage, 1999; Sangketkit et al., 1999). Giang (2010) observed that the calcium oxalate content was reduced 2.45-fold in taro leaf silage compared to the content in fresh taro leaf (0.31 versus 0.76% on a dry matter basis, respectively). Silage-making may be an option to reduce oxalate content in forage plants. However, little is known about the effects of wilting, hay-making, or ensiling on oxalate content in forage plants.

#### CONCLUSIONS

It is clear from the literature that the oxalate content of forage plants depends on many internal and external factors. No single factor can be used to control oxalate content in forage plants. Instead, a large number of factors including fertilizer management, forage species, harvesting practices and seasonal growth should all be taken into consideration. To avoid excess levels of oxalate accumulation, application of nitrate as a sole source of N should be avoided and application of K should be kept to a necessary minimum. It is desirable to use harvesting practices to reduce the oxalate content in some forage plants to acceptable levels, because oxalate levels decline as the harvest interval increases. Forage species or cultivars having a lower tendency to accumulate oxalate should be selected for cultivation. Although oxalate content in forages adversely affects nutritional quality as animal feed, studies on oxalate in forage plants have received little attention. Extensive research is required to understand how plant nutrients minimize oxalate accumulation in plants, through mechanisms at the molecular level. Although application of manure contributes greatly to improving the yield and the nutritional quality of forage, information regarding its effect on oxalate content is limited. The effects of postharvest treatments such as ensiling, wilting and hay-making on oxalate levels are largely unknown. To avoid excess accumulation of oxalate and its harmful effects on livestock, more research is needed to enhance our knowledge of the mechanisms of oxalate accumulation in forage plants.

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