



Assessment of Hydrocyanic Acid (HCN) Content during Different Stages of Growth in Johnson Grass (*Sorghum helepensis*)

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ABSTRACT

Background: Johnson grass (*Sorghum helepensis*) is not a cultivated fodder crop but it is a wild variety seen on farm bunds after rainfall in which *Sorghum vulgare* (Gundrijowar) fodder crops were cultivated in kharif season in Saurashtra region of Gujarat state. At different stages of growth, Johnson grass contains cyanogenic glycosides of varying level which liberate hydrocyanic acid in rumen when it consumed by ruminant animals due to rumen microflora (Toxic level of HCN is 20 mg/ 100gm dry matter). The current study aimed to estimate varying levels of HCN in Johnson grass to keep dairy farmers informed of safe level of HCN for harvesting.

Methods: An investigation was carried out at Cattle Breeding Farm, Junagadh Agricultural University, Junagadh, Gujarat for three consecutive years 2013-2015 in kharif season. Non-cultivated (*Sorghum helepensis*) fodder samples were collected for quantitative HCN concentration from farm bunds nearby eight sown plots of Gundrijowar (*Sorghum vulgare*) from 15 DAS and sampling were continued at fortnightly intervals till the plots exhibit 25% flowering stage.

Result: Result of obtained data in present study revealed that concentration of HCN in Johnson grass in kharif season decreased significantly ($p < 0.05$) on attaining 25% flowering stage during the year 2013, 2014 and 2015. It was concluded that Green Johnson grass can be fed to ruminant animals safely at 25% flowering stage.

Key words: DAS (Day of sowing), Gundrijowar (*Sorghum vulgare*), HCN, Johnson grass (*Sorghum helepensis*).

INTRODUCTION

Johnson grass (*Sorghum helepensis*) available on farm bunds as a unconventional fodder and it is nutritious, palatable and can be fed as green or dry fodder to animals. Cyanide occurs in the leaves of Johnson grass as cyanogenic glycoside and dhurrin. Degradation of dhurrin yields equimolar amount of hydrocyanic, glucose and P-hydroxybenzaldehyde (P-HB). Srinivasa *et al.* (2006) and AL-Sultan (2003) reported high HCN content in the sorghum plant in early growth stage, which decreased with plant maturity. It is observed that when HCN is readily absorbed into the blood stream of grazing ruminants, it causes cellular asphyxiation leading to illness of cattle eventually resulting in the death sometimes. Present experiment was planned to estimate varying levels of HCN in Johnson grass to keep dairy farmers informed of safe level of HCN for harvesting.

It has observed that when HCN is readily absorbed into the blood stream of grazing ruminants, it causes cellular asphyxiation leading to illness of cattle eventually resulting in the death of animals and even at doses as little as 0.5 g are sufficient to kill a cow. The safe limit of HCN in green forage for livestock is 500 ppm on fresh weight basis and 200 ppm on dry weight basis (Karthika and Kalpana, 2017).

MATERIALS AND METHODS

Location of study

An investigation was carried out at Cattle Breeding Farm, Junagadh Agricultural University, Junagadh during the year

Cattle Breeding Farm, Junagadh University, Junagadh-362 001, Gujarat, India.

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2013-2015 in kharif season. Cattle Breeding Farm, Junagadh is located at 70.5° west longitude and 21.4 0 north latitude and is about 60 meters above mean sea level. The climate of area is mainly tropical to sub-tropical and weather parameters (temperature, relative humidity, rainfall, wind speed and sunshine) during year 2013-2015 are presented in Table 1.

Sampling materials

Soil samples for Nitrogen, Phosphorus and Potash were done for each selected plots in which Gundrijowar (*Sorghum vulgare*) were cultivated and it is presented in Table 2. Non-cultivated (*Sorghum helepensis*) fodder samples were collected for quantitative HCN concentration from farm bunds nearby eight sown plots of Gundrijowar (*Sorghum vulgare*) from 15 DAS and sampling were continued at fortnightly intervals till the plots exhibit 25% flowering stage.

Design of experiment

The present research trial was conducted on non-cultivated Johnson grass (*Sorghum helepensis*) available at farm bunds of randomly selected 8 plots (100x100 sq meter sizes) in which *Sorghum vulgare* (Gundrijowar) fodder crops were cultivated in *kharif* season from year 2013-2015.

Statistical analysis

All the recorded data were subjected to statistical analysis by "factorial and completely randomized design" (FCRD) employing one-way analysis of variance as per Snedecor and Cochran (1994). A p-value of <0.05 was considered as significant difference among the treatments groups and the comparison of means were tested as per Duncan's multiple range test (DMRT) described by Duncan (1955).

Procedure for quantitative estimation of HCN (mg/100 gm dry matter) concentration in Johnson grass (*Sorghum helepensis*)

Quantitative analysis of HCN was done according to AOAC (1995) as under

- Two gram of green chopped Johnson grass sample was taken in a test tube and moistened with distilled water. Few drops of chloroform were added to the contents of the test tube.

- Sodium picrate filter paper strips (1x10 cm Whatmanno.1) were prepared by dipping the strips in to solution containing 1% picric acid and 10% sodium carbonate and, air dried.
- Filter papers were inserted into the test tube containing fodder sample and closed with a rubber cork.
- Change in color of filter paper happened immediately after five minutes, however strips were allowed to remain for about six hours in the test tube.
- Filter paper turned from yellow color to brick red color as per the concentration of HCN.
- After evaluating qualitatively, filter paper strips were removed from the test tube chopped to small pieces in to another test tube containing 10 ml distilled water.
- Filter paper strips were thoroughly mixed in a Cyclomixer and centrifuged at 5000 rpm for 10 minutes and color intensity was read using spectronic 20 at 520 nm.
- Standard curve was prepared by following the above method using potassium cyanide.

RESULTS AND DISCUSSION

Randomly selected 8 plots (100 x 100 sq meter sizes) in which *Sorghum vulgare* (Gundrijowar) fodder crops were cultivated in *kharif* season from year 2013-2015. In these *Sorghum vulgare* (Gundrijowar) fodder crops concentration of HCN (mg/100 gm dry matter) was decreased significantly

Table 1: Mean monthly weather data during 2013-2015.

Months	Temperature (°C)	Relative humidity (%)	Rainfall (mm)	Wind speed (km/hour)	Sunshine (hour)
January	20.10±0.40	47.60±4.60	0.00±0.00	5.00±0.30	8.2±0.30
February	23.50±0.40	47.30±3.70	0.00±0.00	4.80±0.10	8.9±0.30
March	27.00±0.70	45.00±5.00	0.30±0.20	5.40±0.10	9.4±0.30
April	30.50±0.30	49.20±1.80	0.10±0.10	6.50±0.60	9.6±0.30
May	32.60±0.20	57.10±2.80	0.00±0.00	8.40±0.50	10.3±0.10
June	30.90±0.40	71.40±2.60	11.10±3.40	9.20±1.20	4.3±0.70
July	28.50±0.60	84.70±2.00	10.50±1.20	8.60±0.80	1.2±0.70
August	27.80±0.50	83.00±2.90	5.20±2.70	4.80±0.60	1.6±0.30
September	28.10±0.50	76.50±2.00	10.10±1.50	3.20±0.60	5.5±0.40
October	28.90±0.60	62.50±2.50	1.20±0.80	2.00±0.20	8.0±0.30
November	26.00±0.40	55.10±2.80	0.40±0.40	2.60±0.40	8.4±0.40
December	21.40±0.30	50.30±2.60	0.00±0.00	3.90±0.30	8.4±0.20

Table 2: Soil analysis report of the plots in which Gundrijowar (*Sorghum vulgare*) fodder grown.

Plot no.	(1:2.5)		O.C. %	Nitrogen kg/ha	P ₂ O ₅ kg/ha	K ₂ O kg/ha
	EC ds /m	pH				
11/3	0.28	8.15	1.02 High	355.0 High	39.49 Medium	110.0 Low
11/4	0.25	8.2	0.81 High	389.0 High	35.39 Medium	99.0 Low
12/2	0.37	7.97	0.99 High	317.0 High	50.27 Medium	320.0 High
12/3	0.51	7.93	0.93 High	279.0 Medium	44.62 Medium	192.0 Medium
12/4	0.31	7.96	0.99 High	622.0 High	67.19 High	386.0 High
14/1	0.27	8.45	0.90 High	299.0 Medium	40.01 Medium	82.0 Low
14/2	0.36	8.43	0.99 High	302.0 High	28.21 Medium	148.0 Low
14/3,4	0.33	8.18	0.90 High	540.0 High	37.44 Medium	121.0 Low

From soil composition it is evident that N, P and K concentrations were usual in comparison to other plots in Cattle Breeding Farm.

Table 3: Concentration of HCN (mg/100 gm dry matter) in Johnson grass (*Sorghum helepensis*) at different stages of growth in Kharif season.

Treatment	Kharif-2013	Kharif-2014	Kharif-2015	Pooled
15 DAS	260.50	284.13	277.13	273.92
30 DAS	133.75	148.00	151.25	144.33
45 DAS	85.63	93.88	93.50	91.00
60 DAS	57.63	68.88	65.13	63.88
25% flowering	17.88	20.75	21.00	19.88
S.E.m.±	5.74	6.26	5.58	3.39
C.D. at 5%	16.50	17.99	16.02	9.51
C.V. %	14.62	14.39	12.97	13.99
Year				
S.E.m.±				2.62
C.D. at 5%				7.37
YXT				
S.E.m.±				5.87
C.D. at 5%				NS

($p < 0.05$) up to 25% flowering stage in kharif season during 2013 and 2014, respectively. In the year 2015, the concentration declined significantly up to 60 days of flowering, while concentrations were at par on 60 DAS and 25% flowering. In the year 2015, HCN concentration though declined from 15 DAS to 45 DAS significantly and it was at par at 60 DAS and 25% flowering. The present research trial was conducted on non-cultivated Johnson grass (*Sorghum helepensis*) available at farm bunds of randomly selected 8 plots (100 x 100 sq meter sizes) in which Sorghum vulgare (Gundrijowar) fodder crops were cultivated in kharif season from year 2013-2015.

Concentration of HCN (mg/100 gm dry matter) in Johnson grass (*Sorghum helepensis*) was decreased significantly ($p < 0.05$) with progressive stages of growth up to 25% flowering stage in kharif season during 2013, 2014 and 2015 respectively as well as in pooled data of 3 years. (Table 3).

Gilchrist *et al.* (1967) found that HCN content <200 mg/g on dry-weight basis is safe for animal consumption. Content of HCN was estimated on green plant samples (0.20 g of minced tissue) sampled at 35 days after sowing. Muthuswamy *et al.* (1976) reported that HCN content was more at the early stage of crop and it decreased at maturity stage. They found that the HCN content was high 18 days after sowing and decreased gradually up to 53 days in CSH 5 type of hybrid variety of sorghum.

Wheeler *et al.* (1990) also reported that hydrocyanic acid (HCN) is an anti-nutritional factor which is potentially toxic to the animal when fed on 30–35 day-old sorghum crops. Chaturvedi *et al.* (1994) reported that HCN content decreased significantly from 65 days after sowing to gain maturity stage. Kumar and Devendra (2010) recorded that decreasing trend in HCN content was observed with the advancement in the fodder growth. The difference for the same were highly significant ($P < 0.01$).

Pandey *et al.* (2011) stated that HCN content in sorghum fodder is high when it grown in soil rich with nitrogen and phosphorous. Zahid *et al.* (2012) observed that Sorghum cultivars during rainfed regions produce lowest level of HCN and at pre-booting stage is safer for livestock feeding. Jadav *et al.* (2019) reported that concentration of HCN (mg/100 gm dry matter) in Gundrijowar fodder crop was decreased significantly ($p < 0.05$) up to 25% flowering stage in summer season during the year 2013 and 2015. While in the year 2014 significant decline was up to 60 DAS and the concentration was at par at 60 DAS and 25% of flowering stage. Pooled data showed similar trend to that of the year 2014 with reference to HCN concentration

CONCLUSION

It could be concluded that irrespective of the initial HCN concentration (mg/100gm dry matter) in Johnson grass (*Sorghum helepensis*), it declined to safe level of feeding to ruminant animals at 25% flowering stage.

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