Potential of waste flowers used as feed additive on the performance of goat kids

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The area under floriculture in India during 2007–08 to 2017-18 increased from 0.56 to 1.04 million hectares, while production increased from 1.26 to 3.65 million tonnes (mt), respectively. Out of which only 0.02 mt of flowers worth ₹ 5,073 million were exported in 2017–18 and the remaining were utilized domestically (ARDB 2019). Fresh flowers have been used in worship, decoration, weddings, festivals and funeral rituals, as an intrinsic element of the entire human culture. After the above ceremonies, huge quantities of fresh waste flowers (WFs) are available for disposal. Because of the religious sanctities the WFs cannot be dumped into the garbage, therefore disposed of in rivers or nearby water bodies or dumped in waste land, causing choking of water bodies, foul smell, unhygienic atmosphere and spread of infectious diseases, besides causing pollution.

Edible flowers have been used in various foods and beverages, besides they are known as nutraceuticals because of having phytochemicals and biological properties. Mahindrakar (2018) have reviewed floral waste utilization for various purposes like vermicomposting, conversion to activated carbon, charcoal, extraction of dyes and essential oils and bio-gas generation. Till date no report is available on using WFs as feed additive on ruminant's production. This study was therefore taken up to explore the utilization of WFs as feed additives in the total mixed ration (TMR) on the nutrient utilization and performance of goat kids.

Waste flowers [mixture of marigold (*Calendula officinalis*) and rose (*Rosa indica* L)] procured from local temples were segregated, sun dried and finely ground for analysis. The TMRs supplemented with WFs at 0–5% were evaluated in triplicate by *in vitro* gas production technique (IVGPT, Menke *et al.* 1979). Similar sets were run with 200 mg TMR, supplemented with WFs at 0–5% on dry matter basis for 0, 2, 4, 6, 8, 10, 12, 24, 26, 48, 60, 72, 84, and 96 h to work out $t_{1/2}$ using Graph Pad Prism software (Miller 2003). Another set was run at $t_{1/2}$ to see the fermentation pattern. For CH₄ estimation, representative gas sample was taken from the headspace of syringe in an air-tight 10 μ l Hamilton syringe and injected into gas

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chromatograph. After 24 h incubation, a 5 mL aliquot of fluid from each syringe was mixed with 1 mL of 25% metaphosphoric acid, kept for 1 h at ambient temperature (Erwin et al. 1961) and the Volatile fatty acids (VFAs) were estimated using gas chromatograph (Cottyn and Boucque 1968). Further, fermentation attributes related to hydrogen recovery, VFA utilization index, efficiency of conversion of fermented hexose energy to VFA and efficiency of conversion of fermented hexose energy to methane energy were calculated by adopting procedures described in an earlier publication (Wadhwa et al. 2021).

A 90 days growth trial was conducted on male Beetal goat kids (15 number; 8 months old; average live weight 22.14±1.18 kg) randomly distributed into three equal groups. The animals in control group were fed TMR containing concentrate mixture (maize 32, barley 20, groundnut cake 25, wheat bran 15, full fat soy 5, mineral mixure 2 and salt 1% each) and green oats in 50:50 ratio on DM basis as per NRC (2007) feeding standard for goats. Those in experimental groups were fed control TMR supplemented with dried, ground waste flowers @ 3 or 5% of DM intake. A 7-day digestion-cum-metabolism trial was conducted before the termination of growth trial. During metabolic trial, the animals were kept individually in specially designed metabolic cages where urine and faeces were collected automatically in separate containers.

The finely ground samples of feed, feces and orts were analyzed for CP, EE, total ash (AOAC 2007) and cell wall constituents (Van Soest *et al.* 1991), total phenols (Makkar *et al.* 1993), flavanoids (Balabaa *et al.* 1974), saponins (Baccou *et al.* 1977), anthocyanins (Lapornik *et al.* 2005), 2, 2–diphenyl picrylhydrazyl (DPPH) activity (Kumaran and Karakumaran 2007) and vitamin C (Jagota and Dani 1982). Fresh urine and fecal samples were analyzed for N (AOAC 2007). The data were analyzed by using one way ANOVA (Snedecor and Cochran 1994) by using SPSS (2009) version 16.0 and the means were tested for the significant difference by using Tukey's-b test.

Marigold flower constituted the major proportion of the waste flowers collected, with negligible quantity of rose flowers. The data revealed that marigold flowers had 4.1 times higher (P<0.01) total phenols and 1.6 times higher (P<0.01) flavonoids, while rose had higher (P<0.01) amount

Table 1. Bio-active components in waste flowers (mg %)

Component	Marigold	Rose (red)	PSE	P value	
Total phenols	9.44 ^b	2.29 ^a	3.87	< 0.001	
Anthocynins	0.09^{a}	$0.87^{\rm b}$	0.22	< 0.001	
Saponins	0.14^{a}	0.28^{b}	0.04	< 0.001	
Vitamin C	3.02 ^a	3.90^{b}	0.26	0.002	
Flavonoids	6.16 ^b	3.88^{a}	0.66	0.002	
DPPH	54.55 ^a	55.44 ^b	0.26	0.031	

DPPH, 2, 2-diphenyl picrylhydrazyl; Figures with different superscripts in a row differ significantly.

of anthocynins, saponins, vitamin C by 9.7, 2.0 and 1.3 times respectively than that in marigold (Table 1). The DPPH activity was also higher (P<0.05) in rose flowers than that observed for marigold. Marigold flowers (*Tagetes erecta* L) are a rich source of lutein, a carotenoid pigment (Jain 2017), saponins, triterpenic alcohols (Giuliano *et al.* 2008) and polyunsaturated fatty acids (Kishimoto *et al.* 2005).

The TMR contained 9.09% total ash, 90.92% OM, 11.55% CP, 60.08% NDF, and 27.23% ADF, 21.55% cellulose and 32.85% hemicelluloses on DM basis. The results revealed that Net gas production (NGP), partitioning factor and availability of ME content was not affected by

supplementation of waste flowers, however the digestibility of NDF and that of true OM was improved (P<0.05) by supplementation of waste flowers at 3 and 5% levels (Table 2).

The data revealed that NGP at t½ increased linearly (P<0.01) as the level of supplementation of waste flowers increased in TMR. On an average the NGP increased by 10.8% on supplementation of waste flowers. Methane emission as per cent of NGP at t½ decreased (P<0.01) linearly as the level of WFs supplementation to the TMR increased.

The effect of supplementation of waste flowers to TMR on the fermentation pattern revealed that as compared to control TMR, supplementation of waste flowers at 3 and 5% levels on DM basis resulted in higher (P<0.01) production of total VFAs, acetate and propionate. The flow of hydrogen is diverted into propionate production via lactate or fumarate (Asanuma *et al.* 1999). It was observed that at lower levels of supplementation of WFs the fermentation efficiency was not affected, but it increased (P<0.01) with increase in level of supplementation from 3% onwards. Efficiency with which hexose energy is converted to VFA energy was highest (P<0.01) when the diet was supplemented with waste flowers @ 5%. Energy loss through methane was reduced (P<0.01) with the

Table 2. Impact of supplementing waste flowers to the TMR on NGP, digestibility of nutrients, ME availability, total and individual volatile fatty acids production and hydrogen balance by IVGPT

Parameter	Level of waste flowers, %						PSE	P value
	0	1	2	3	4	5		
NGP, ml/g DM/24 h	180.0	182.61	182.69	183.81	182.05	183.28	0.95	0.168
NDFD, %	26.48a	29.11 ^{ab}	33.54bc	36.83 ^c	32.90^{bc}	31.88bc	1.04	0.005
TOMD, %	58.82a	59.92 ^{ab}	61.51 ^{abc}	63.68 ^c	62.10^{bc}	62.34 ^{bc}	0.85	0.006
PF, mg/ml	2.00	2.04	2.05	2.04	2.07	2.08	_	0.107
ME, MJ/kg DM	8.20	8.28	8.25	8.32	8.28	8.31	_	0.596
NGP and methane prod	duction at t½							
NGP, ml/g DM	125.00 ^a	129.17 ^b	136.67 ^c	138.33 ^c	144.17 ^d	144.17 ^d	1.72	< 0.001
CH ₄ , %	45.18 ^b	43.42 ^{ab}	41.71 ^a	42.72 ^{ab}	40.65 ^a	40.84 ^a	0.82	0.010
VFA production, mM/d	!L							
TVFA	4.93 ^a	4.94 ^a	5.08 ^b	5.33 ^d	5.12 ^b	5.27°	0.04	< 0.001
Acetate (A)	3.55 ^a	3.56 ^a	3.70 ^b	3.83^{d}	3.68^{b}	3.79 ^c	0.03	< 0.001
Propionate (P)	0.849^{a}	0.855a	0.863a	0.93 ^c	0.878^{b}	0.926 ^c	0.01	< 0.001
Isobutyrate	0.049^{a}	0.048^{a}	0.051 ^b	0.052^{b}	0.055^{c}	0.051^{b}	0.001	< 0.001
Butyrate	0.355^{a}	0.355a	0.353a	0.388^{c}	0.378^{b}	0.379^{b}	0.004	0.002
Isovalerate	0.082^{b}	0.077^{a}	0.077^{a}	0.081 ^b	0.082^{b}	0.075^{a}	0.001	< 0.001
Valerate	0.044^{a}	0.045a	0.045a	0.051 ^b	0.054^{c}	0.045^{a}	0.001	< 0.001
A:P	4.10 ^a	4.16 ^b	4.29 ^c	4.11 ^a	4.19 ^b	4.19 ^b	_	< 0.001
Hydrogen balance, %								
E	72.22 ^a	72.02 ^a	72.20 ^a	72.27 ^{bc}	72.24 ^b	72.33°	0.53	< 0.001
E_1	71.91 ^{bc}	71.72 ^a	71.88 ^b	71.97 ^{cd}	71.93 ^{bc}	72.03 ^d	0.44	< 0.001
E_2	17.94 ^a	18.19 ^c	18.04 ^b	17.92 ^a	17.97 ^{ab}	17.89 ^a	0.18	< 0.001
HR	29.09 ^{bc}	28.49 ^a	29.00 ^b	29.21 ^{bc}	29.09 ^{bc}	29.33°	0.11	0.001
HC	0.128ab	0.132 ^c	0.129 ^b	0.127^{ab}	0.128ab	0.126a	0.001	0.001
VFA-UI	4.81 ^b	4.90 ^c	4.83 ^b	4.78 ^b	4.79 ^b	4.73 ^a	_	< 0.001

E, Fermentation efficiency; E_1 , Efficiency of fermented hexose energy to VFA energy; E_1 , Methane energy; HR, Hydrogen recovery; HC, Hydrogen consumed; VFAUI, Volatile fatty acid utilization index. Figures with different superscripts in a row differ significantly.

increase in level of WFs from 1 to 5%.

Acetate and butyrate promote CH₄ production, while propionate formation is for diverting hydrogen away from methane production in the rumen. One mole H₂ is required per mole of propionate or valerate produced. This suggests that the proportions of acetate, butyrate and propionate would determine the amounts of available H₂ for methanogens. Hydrogen balance revealed that hydrogen recovery was highest (P<0.01), while hydrogen consumed was lowest (P<0.01) when the diet was supplemented with 5% waste flowers. The VFA utilization index varied (P<0.01) from 4.73 (diet supplemented with 5% waste flowers) to 4.9 (diet supplemented with 1% waste flowers).

Based on the results of *in vitro* studies, waste flowers at 3 and 5% level were selected for conducting a 90 days feeding trial on goat kids. Supplementation of WFs at 3% did not have any adverse effect on the digestibility of proximate and cell wall constituents as compared to control TMR (Table 3). But, the digestibility of DM, OM, and CP was depressed (P<0.05) at 5% supplementation of WFs as compared to control group. The digestibility of cell wall constituents except hemicelluloses was not affected even at 5% WFs supplementation to the TMR. The nitrogen utilization was not affected by supplementation of waste flowers to the TMR.

The performance of kids was better when diet was supplemented with waste flowers (Table 3). The body weight gain and average daily gain was 14 and 41%

Table 3. Impact of supplementing waste flowers to the TMR on DM intake, digestibility of nutrients, N-retention and gain in weight of goat kids

		5 8			
Parameter		of waste f OM inta			
	0	3	5	PSE	P value
DM intake, g/day	780	840	850	60.0	0.755
Digestibility of nu	trients,	%			
DM	67.82 ^b	61.90 ^{ab}	60.72a	1.29	0.035
OM	71.10^{b}	64.78ab	63.75a	1.37	0.040
CP	81.28 ^b	74.87^{ab}	72.25 ^a	1.58	0.045
NDF	63.85	56.16	56.03	1.66	0.074
ADF	36.34	32.93	30.18	1.70	0.365
Cellulose	57.60	47.59	51.75	2.03	0.123
Hemicellulose	82.44 ^b	75.32 ^{ab}	73.21 ^a	1.69	0.047
N-retention, g/day	,				
N-intake	19.23	20.40	20.58	1.21	0.758
Faecal-N	2.94	3.90	4.60	1.14	0.116
Urinary-N	3.90	4.43	5.60	0.55	0.078
N-retained	12.39	12.07	10.38	0.96	0.313
Gain in weight					
Initial BW, kg	22.07	22.2	22.15	1.18	0.999
Final BW, kg	27.32	28.21	29.58	1.37	0.819
BW gain, kg	5.27	6.01	7.43	0.53	0.251
ADG, g	58.59	66.74	82.52	5.85	0.251

Figures with different superscripts in a row differ significantly.

respectively at 3 and 5% level of supplementation of waste flowers in the diet.

SUMMARY

The present study was conducted to assess the impact of temple waste flowers as feed additive on the growth of goat kids. Marigold (Calendula officinalis) flowers constituted the bulk of WFs with negligible red roses (Rosa indica L). Marigold flowers as compared to rose flowers contained high (P<0.01) total phenols and flavonoids; and low (P<0.01) anthocyanins, saponins and vitamin C content. The in vitro studies revealed that the supplementation of WFs at 3 and 5% improved (P<0.01) digestibility of nutrients and VFAs production as compared to control. NGP at t1/2 increased (P<0.05) linearly up to 4% WFs supplementation and methane production decreased (P<0.05) at all levels of supplementation. Fifteen Beetal goat kids divided into three equal groups were fed TMR, or TMR supplemented with WFs at 3 or 5% of DM intake for 90 days. The daily DM consumed by the animals was similar in all the groups. The digestibility of proximate constituents at 3% level of WF was comparable with control TMR, but depressed (P<0.05) at 5% level of supplementation. The digestibility of cell wall constituents and N retention were not affected by WF supplementation. The average daily gain in weight was improved considerably. It was concluded that bio-active compounds present in WFs reduced the methane emission, resulting in improvement in nutrient utilization and growth of goat kids.

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