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Effect of inclusion of subabul tree fodder on the physical properties and nutritive value of maize silage

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Abstract
Subabul (*Leucaena leucocephala*), a leguminous tree fodder having protein levels between 27 percent to 34 percent and its adaptability to tropical climates, plays a crucial role by providing sustainable nutrition for cattle, goats, and other livestock, especially during tough environmental periods such as droughts. A study was conducted to evaluate the physical characteristics and nutritive value of maize silage with different levels of subabul tree fodder (0, 10, 20 and 30 percent). After six weeks of ensiling, the maize-subabul mixed silage was opened and examined for physical characteristics and nutritional values. All combinations of silage were yellowish-green in colour with a sweet aroma and pH ranging from 3.79 to 4.33 with no signs of fungal growth. Three to seven percent decrease in dry matter content was observed in the respective silage samples of all the treatment groups. The silages were analysed for their crude protein, ether extract and total ash contents. The protein content was significantly ($p<0.05$) increased in treatments with a higher percentage of subabul fodder. In both fresh fodder and silage, the CP content was significantly ($p<0.05$) higher in the M+S (70+30) group. The EE content was significantly ($p<0.05$) increased for all the treatment groups and the TA content was significantly ($p<0.05$) decreased for all groups except M+S (70+30) group after ensiling. The study demonstrates that silages prepared using legume tree fodder have desirable physical characteristics and nutritional values and high-quality protein-rich silages can be prepared using legume fodders up to 30 percent level of inclusion.

Keywords: Ensiling, mixed silage, crude protein, ether extract and total ash

Introduction
In the Indian agricultural landscape, where the demand for high-quality fodder is ever-increasing due to the burgeoning livestock industry, sustainable and efficient forage production practices are paramount. Ensuring the consistent year-round availability of green fodder has been identified as a major constraint in optimizing the economic efficiency of dairy cattle production in India [13]. The total amount of green fodder available in India was calculated to be 734.2 million metric tons, falling short of the required 827.19 million metric tons by 11.24% [20]. Improving the quality of the available fodder and conserving it efficiently is the most important thing to consider in the current Indian scenario. Silage-making has gained prominence recently as a preservation method, presenting a viable solution to the challenges associated with seasonal variations in fodder availability. According to [22], ensiling stands out as a cost-effective technique for preserving fodder, which can aid in fulfilling the demand for green fodder during periods of scarcity. Under the fermentation process, the growth of harmful microorganisms can be inhibited and nutrient content can be preserved [23]. Leguminous tree fodder plays a vital role in Indian livestock production by providing a sustainable nutritional source for cattle, goats, and other livestock, particularly during challenging environmental conditions like droughts. Among other fodder trees, Subabul (*Leucaena leucocephala*), stands out for its high nutritional value having protein content ranging from 27 to 34 percent, along with significant levels of carotene and vitamin A and its ability to thrive in tropical climates thereby contributing significantly to livestock feed [11]. However, tropical grasses and legumes are not naturally conducive to ensilage due to their low water-soluble carbohydrate levels at harvest and their high buffering capacity, which hampers pH changes.

Despite these challenges, several techniques like wilting, combining legumes with cereal crops, incorporating silage additives, and utilizing small-scale silos can enhance fermentable carbohydrate levels, reduce buffering capacity and inhibit proteolysis of leguminous fodder thus ensuring the production of high-quality protein-rich silage.

Materials and Methods

Experimental Details: The research was conducted at the Department of Livestock Production Management, Veterinary College, Hebbal, Bengaluru to evaluate the physical characteristics and nutritive value of maize silage with different levels of subabul tree fodder. The laboratory silages for the study were prepared using the combinations of one cereal fodder (maize - 100, 90, 80 and 70%) and one legume fodder (subabul – 0, 10, 20 and 30%) in triplicates. Whole maize fodder at the milk stage and subabul tree fodder were harvested, wilted and chopped to 2-3 cm size for making the silage. The chopped maize and subabul fodder were mixed in a ratio of 90:10 by adding 10 kg subabul fodder to 90 kg Maize fodder and mixing it thoroughly by spreading it on a polythene sheet to a depth of 10 cm. Similarly, 80:20 (80 kg maize and 20 kg subabul), 70:30 (70 kg maize and 30 kg subabul) and 100:0 (100 kg maize) combinations were also mixed separately. The silages were filled in 10 litre fibre/plastic containers according to normal silage preparation techniques and sealed for six weeks at room temperature [6]. After six weeks, the plastic silo containers were opened, and laboratory analysis was performed with samples collected from the treatments each with three replicates.

Table 1: Design of experiment with treatments and different proportions of legume and non-legume fodder varieties used for the study

Treatment	Sample name	Maize %	Subabul %
T1 (Control)	M	100	0
T2	M+S (90+10)	90	10
T3	M+S (80+20)	80	20
T4	M+S (70+30)	70	30

Evaluation of physical properties and nutritive value: The odour and colour of the silage was recorded as per [16]. After opening the lid, the silage was examined for the presence or absence of other extraneous materials like mold growth or worms [6]. The pH of silages was estimated by the method described by [19]. The processed silage samples were analysed for proximate principles *viz*., crude protein, ether extract and total ash as per the standard methods described by the AOAC [3] and the fibre fractions *viz*., neutral detergent fibre, acid detergent fibre and acid detergent lignin were determined as per the method described by [24]. The toluene dry matter (DM) of ensiled material was calculated using the formula Toluene DM = (0.95 Oven DM + 3.3) [8].

Statistical analysis: All the data of various parameters were analysed statistically by one-way ANOVA and two-way ANOVA using R statistical software. Differences between the means were tested using Tukey's test at $p<0.05$.

Results and Discussion

Physical Properties: The colour, smell, visibility of mold growth, pH and dry matter of silages prepared using maize fodder with subabul inclusion at different levels was recorded after six weeks of ensiling. The colour of all combinations of silages was yellowish-green, which is suggestive of very good quality silage. The results agree with the findings of [17] and [7]. All the silage combinations had sweet aromas and no fungal growth was noticed in any of them which is indicative of good-quality silage agreeing with the observations of studies by [1] and [2]. The pH of the silages of maize with different levels of subabul tree fodder ranged from 3.79 to 4.33, which was consistent with the results reported by [17] and [1]. As the proportion of legume fodders to cereals increased, so did the pH value because legumes have a low content of fermentable carbohydrates required for lactic acid bacteria (LAB) fermentation, a high crude protein content, and resistance to pH change due to the high buffering capacity of legumes [27]. This was consistent with a study conducted by [21], in which pea grass was employed as the legume in a mixture with cereal.

The dry matter content of the fodder used for silages of maize with different levels of subabul tree fodder without bacterial inoculum ranged from 24.80 and 28.61 percent and that of the silages ranged from 23.83 and 27.05 percent. A significant ($p<0.05$) increase in dry matter content was observed in silages with 20 percent and 30 percent inclusion of subabul tree fodder. The lesser moisture content of subabul fodder when compared to maize fodder, might be the reason for the increase in DM content of the fodder and silage samples with higher levels of subabul addition. Comparable results were observed by [1] when maiwa millet (*P. glaucum* L.) was ensiled with the trailing, annual legumes and contrary to [4] when sweet sorghum was replaced with a higher percent of lucerne for ensiling. Three to seven percent decrease in dry matter content was observed in respective silage samples of all the treatment groups but the change was not significant. Microorganisms can break down easily degradable silage ingredients (e.g., WSC) into silage acids, ethanol, and carbon dioxide during fermentation [4]. [14] reported that loss of nutrients in silage is through oxidations that take place in normal fermentation and other changes occurring in silage fermentation process.

Table 2: Physical properties of maize silages prepared by the addition of different levels of subabul tree fodder

Parameter		M	M+S (90+10)	M+S (80+20)	M+S (70+30)
Colour		Yellowish green	Yellowish green	Yellowish green	Yellowish green
Smell		Sweet aroma	Sweet aroma	Sweet aroma	Sweet aroma
Mold growth Visibility		Nil	Nil	Nil	Nil
DM %	Fodder	24.80 ± 0.42^{B}	26.14 ± 0.14^{B}	26.28 ± 0.30^{B}	28.61 ± 0.26^{A}
	Silage	23.83 ± 0.28^{B}	24.8 ± 0.19^{B}	25.07 ± 0.31^{BC}	27.05 ± 0.23^{AC}
pH		3.79 ± 0.00^{C}	4.00 ± 0.04^{B}	4.09 ± 0.03^{B}	4.33 ± 0.03^{A}

Means in the same row with no common superscripts differ significantly ($p<0.05$)

Nutritive Value: The mean CP content of the fresh fodders ranged from 6.46 to 14.78 percent and that of the silages ranged from 5.97 to 13.54 percent. The protein content was significantly (p<0.05) increased in treatments with a higher percentage of subabul fodder. In both fresh fodder and silage, the CP content was significantly (p<0.05) higher in the M+S (70+30) group and lower in the M group when compared to other treatment groups. Legumes have a higher protein and lower fibre content than maize, leading to an increase in CP content in legume-mixed silages [15]. [18, 4, 10] reported similar findings when legumes were added to cereal fodders in varying proportions. The CP content of the treatments was reduced after ensiling. This lower CP in the silage compared to fresh fodders could be due to protein degradation during ensiling, which might have resulted in more non-protein nitrogen in the silage than in the herbage before ensiling [26]. The current investigation yielded results comparable to those reported by [18, 4].

In the treatment groups of silages of maize with subabul in different proportions the ether extract was significantly (p>0.05) decreased in treatments with a higher percentage of subabul for fresh fodder samples meanwhile there was no significant (p<0.05) change in silage samples. The findings obtained in this study are in agreement with the results of [4, 5] in which the inclusion of legume for ensiling did not affect the concentration of EE. The EE content of all the treatments was significantly (p<0.05) increased after ensiling. It was significantly (p<0.05) higher in the silage samples of all treatments. The possible reason could be that microorganisms, particularly LAB contribute to the breakdown of plant cell walls, releasing intracellular lipids, and influencing the ether extract content [25].

In both fresh fodder and silages of the treatment groups of maize with subabul in different proportions with and without the addition of bacterial inoculum, the total ash content was significantly (p<0.05) increased in treatments with a higher percentage of subabul fodder. The TA content was significantly (p<0.05) higher in the M+S (70+30) groups and lower in the M groups when compared to other treatment groups. Legumes have higher ash content than cereal fodders, leading to an increase in total ash content compared to the control group. Similar findings were reported by [4, 5]. The TA content of the treatments was significantly (p<0.05) decreased after ensiling. It was significantly (p<0.05) higher in the fresh fodder samples when compared to the respective silages of all the treatments. Since the ash concentrations in silage effluent are higher than those in herbage DM, a decrease in the ash concentration in the silage has been attributed to losses in silage effluent [12]. The outcome was comparable to the findings reported by [9, 10, 18].

Table 3: Proximate analysis of fresh fodder versus laboratory silage of maize and subabul in different proportions

Treatment and Parameter		M	M+S (90+10)	M+S (80+20)	M+S (70+30)
CP %	Fresh Fodder	6.46±0.07^{D}	9.49±0.24^{C}	11.87±0.10^{B}	14.78±0.06^{A}
	Silage	5.97±0.02^{D}	8.41±0.28^{C}	11.12±0.22^{B}	13.54±0.27^{A}
	P - value	0.5718	0.0082	0.7512	0.2443
EE %	Fresh Fodder	2.43±0.15^{A}	2.1±0.03^{AB}	1.86±0.06^{B}	1.14±0.04^{C}
	Silage	3.49±0.03^{A}	3.45±0.03^{A}	3.40±0.02^{A}	3.12±0.04^{A}
	P - value	<0.0001	<0.0001	<0.0001	<0.0001
TA %	Fresh Fodder	6.63±0.02^{D}	6.98±0.02^{C}	7.23±0.02^{A}	7.45±0.06^{AB}
	Silage	6.35±0.01^{C}	6.61±0.01^{B}	6.72±0.06^{B}	7.25±0.13^{A}
	P - value	0.0005	<0.0001	<0.0001	0.9763

*Mean in the same row with no common superscript differs significantly (p≤0.05).
*Level of significance – 5%
*CP-Crude Protein, EE-Ether extract, TA-Total Ash

Conclusion

In conclusion, the study demonstrates that silages prepared using legume tree fodder have desirable physical characteristics and nutritive values. Even though there was a slight increase in the pH of the silage with 30 percent addition of subabul fodder, the remaining physical characteristics and nutritive values were favourable. There was a significant increase in the crude protein content of silages with higher levels of subabul. The study suggests that high-quality protein-rich silages can be prepared using legume fodders up to 30 percent level of inclusion.

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