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# Fractionation of the Foliages Obtained from Leguminous Species for Leaf Protein Preparation

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#### **ABSTRACT**

The process of green crop fractionation (GCF) has been recommended for producing high quality feed and food grade products for animal and human consumption respectively. The pressed crop (PC) residue released during fractionation is considered as a feed product which is fibrous in nature containing adequate nutrients to fulfil the requirements of cattle. This product is more digestible than whole unfractionated crop. The digestibility of PC residue increases due to the mechanical treatment i.e. maceration. Most of the proteins are extracted in juice and therefore the protein content in PC residue is less, in comparison to that in unfractionated foliage. Furthermore during pressing a large amount of moisture is released in the form of juice. The PC residue is thus low moisture product with low protein content. This character of PC enables to make good quality of silage. The leaf protein concentrate (LPC) obtained during GCF is free from indigestible fibre and it contains high proportions of proteins which are nutritionally superior. For this reason, its use in the nutrition of human beings and non - ruminants has been advocated. Apart from proteins, it contains appreciable amounts of vitamin A, minerals, fats and vitamin E. Studies of various places in India and abroad indicated its suitability in human nutrition.

Three crops, i.e., Vigna, cowpea and Dolichos were selected to find out its production potential before and after fractionation. These were earlier found suitable for fractionation and have been recommended for LP production. It is indicated that the crops are highly productive and can be raised without much use of fertiliser. As all the three crops were leguminous they would fix nitrogen and save fertilizer input. The data indicated that all the three crops produced appreciable amounts of PC residue as well as LPC and these crops could be employed for commercial production of leaf protein and fibrous residue. Their comparison with lucerne revealed that as these crops as annual; they could produce the products only during particular season. Hence detail investigation and crop sequence studies are required to produce LPC round the year from such suitable crops.

Keywords: Green crop, Vigna, yield, Pressed crop, Dry matter, Protein

## INTRODUCTION

The yields of PC, LPC and DPJ: Several types of crops have been recommended from GCF (green crop fractionation) by Pirie [1] and LP production [2-5]. The squeezed pulp when obtains the juice, it is heated at 90°C to precipitate LPC [6-9]. This LPC (Leaf Protein Concentrate) when isolated by filtration, it gives the filtrate liquid residue called as Deproteinised Juice (DPJ). For GCF include fodder crops, cover crops, vegetables green manuring crops and by-product leaves of some vegetables [10].

Earlier investigations on agronomic aspects of L P production have shown that lucerne berseem is the most promising species for green crop fractionation [11-15]. Mung (*Vigna radiata* Wilzech) is a common pulse crop in this region. It grows well in wet season and in some part of the country, its foliage is used as fodder. It is an admirable crop for leaf protein extraction during present investigation, this crop was cultivated harvested and fractionated in order to measure its production potential [16-17].

Cowpea (*Vigna unguiculata* (L.) Walp) is mainly cultivated as a vegetative crop in this region for young green pods [18-24]. Earlier studies by Deshmukh [25] with crop at this centre revealed that from 40-63% of protein can be

extracted from this crop. Mungikar [22] cultivated cowpea during monsoon when the rate of LP production was 3.2 to 3.8 kg per hectare per day. Dakore [24] cultivated this crop as an intercrop with either maize or Sorghum and pointed out that it thrives well with Sorghum. During present study this crop was cultivated for the measurement of its yields. *Dolichos lablab* is also another important leguminous crop in this region. It is also cultivated for edible pods as a vegetable crop. In rural areas it is cultivated as either a green manure or around the fields of sugarcane. Deshmukh [25] recorded that the crop can yield upto 70 q of green fodder per hectare. Basole [26] and Bhuktar [27] cultivated Dolichos in association with either Sorghum or maize fields. During present study this crop was cultivated to observe its fractionation characteristics [28]. The main objective in undertaking present study was to evaluate the performance of Vigna, *Dolichos* and Cowpea for fractionation and to measure the yields of various fractions [29].

#### MATERIALS AND METHODS

#### Agronomy

The field trial was conducted at Dr. Babasaheb Ambedkar Marathwada University Botanical Garden. The soil at the experimental site was slightly alkaline with a pH value of 8.2 containing 0.45 to 0.62% organic carbons (O.C.). The available N, P and K were 12.4, 12.2 and 24.0 per hectare, respectively. A piece of land was prepared by ploughings and cross ploughings. The land was then divided into plots, each bearing 16.48 m² areas. The three crops viz. Vigna, Cowpea and *Dolichos* were sown on 29.7.1995. The seed rates used were Rs. 35, 50 and 15 kg/ha, respectively for Vigna, Cowpea and *Dolichos*. The sowing was done in rows, 30.5 cm apart and a basal dose of 20 kg N, 20 kg P and 10 kg K in combination with farmyard manure (FYM) was applied at the time of sowing. Since the crops were to be harvested for green fodder, slightly higher seed rates were used than those recommended for the production of grains. The crops were grown under irrigation and as far as possible the use of insecticides and pesticides were avoided. The field trial was conducted in randomised block design (RBD) with three replicates. The green foliage from each crop was harvested at a pre-flowering stage, 48 days after sowing.

## Sampling

The crops were harvested for green fodder with a steel cutter early in the morning. As mentioned earlier, the harvesting was usually done at a pre-flowering stage. Fresh green fodder obtained per plot was measured and recorded as field of green fodder. A sample of green fodder obtained after harvesting from each crop was immediately brought into the laboratory for fractionation.

## Fractionation

3 kg of green fodder was pulped using IBP pulper. A sample of the pulp was taken for the determination of dry matter. 900 g pulp was pressed on laboratory scale IBP press for 10 min. The amount of juice expressed due to the pressing was measured and a sample of the juice was taken for preparation of leaf protein concentrates. The pressed crop residue or fibrous residue, left after the extraction of juice from 900 g pulp was weighed and sample was taken for the estimation of dry matter (DM).

For the preparation of LPC, about 20 ml of water was heated in a beaker allowed to boil. 50 ml of juice was slowly added to the boiling water with stirring and the contents were heated 90-95°C, when proteins in juice coagulated resulting into a curd of LPC. The heated juice was then filtered through a cotton cloth and the weight of LPC was recorded. A sample of LPC was taken for the determination of dry matter.

The samples of pulp, pressed crop (PC) and LPC were dried in oven at  $90 \pm 5$ °C for 48 h till constant weight for the determination of percent dry matter (DM). The dried samples were ground to a fine powder and used for the estimation of nitrogen.

Nitrogen (N) content was determined by Microkjeldahl method with sulphuric acid (H<sub>2</sub>SO<sub>4</sub>) in presence of catalyst (9 K<sub>2</sub>SO<sub>4</sub>:1 CuSO<sub>4</sub>:0.02 SeO<sub>2</sub>) and titration of ammonia liberated

During distillation [30], the value of crude protein (CP) was expressed as  $N \times 6.25$ . The yields of green foliage, dry matter and crude protein on the field and that obtained for pressed crop and LPC were calculated with the help of amount of green fodder harvested per plot , amount of pressed crop and juice obtained per 900 g pulped foliage, amount of LPC obtained per 50 ml of juice, the DM content of pulp, PC and LPC and the N content in these 3 fractions. The data was analysed for analysis of variance (ANOVA) and the critical difference was calculated following Panse and Sukhatme [31].

## RESULTS AND DISCUSSION

All the three crops grew well with abundant green foliage. The yield of green fodder/hectare was highest in Vigna 7173 kg/hectare followed by cowpea and *Dolichos* which yielded 5181 kg/hectare green fodder. The results are in agreement to those obtained during earlier field trials in this laboratory. The dry matter contents in the foliages of Vigna, Cowpea and *Dolichos* were 20.2, 14.8 and 19.6%, respectively. The yields of DM reached 1483, 758 and 1019 kg/hectare respectively in Vigna, cowpea and *Dolichos*. The N % of dry matter in the three crops ranged between 2.78 and 2.84. Nitrogen content in the foliage was lower than that recorded earlier probably due to very limited use of fertilizers. The yields of crude protein is reached 259 in Vigna, 133 in cowpea while 177 in *Dolichos*.

The overall results obtained and presented in Table 1 and Figures 1A and 1B indicated that the three leguminous crops are suitable for production of green foliage. Higher yields of leaves from this crop as well as high nitrogen content in the foliage could be obtained by using fertilizers as was experienced earlier in this laboratory. The green foliages from three crops were subjected to fractionation using IBP equipments. The main aim was to measure the yields of pressed crop which is suitable for animal nutrition and that of leaf protein concentrate, the use of which is recommended the nutrition of non-ruminants including calf, poultry and human beings. Table 2 gives data on the crop residues while Table 3 on that of leafy protein concentrates obtained due to the fractionation of three crops.

Table 1: The yields of green fodder, dry matter and crude protein (all in kg/ha) from Vigna, cowpea and Dolichos

Crop	Green Foliage	Green Foliage		Yields (kg/ha )			
	% Dry matter	N % of DM	Green fodder	Dry matter	Crude protein		
Vigna	20.2	2.84	7173	1483	259		
Cowpea	14.83	2.78	5181	758	133		
Dolichos	19.66	2.8	5181	1019	178		
Mean	18.23	2.8	5845	1086	190		
Std. Deviation	2.95	0.03	1150	367	63.85		
Std. Error	1.7	0.01	664	212	37		
C.V.	16.18	1.07	19.67	33.79	33.6		

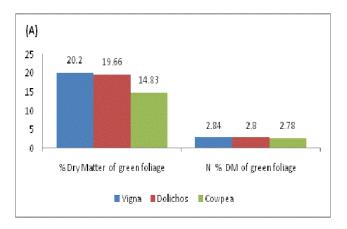


Figure 1A: The yields of percent dry matter of green foliage and percent nitrogen content

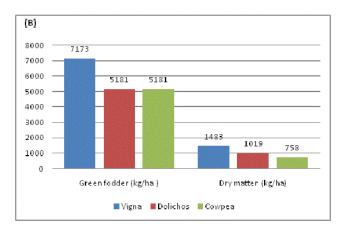
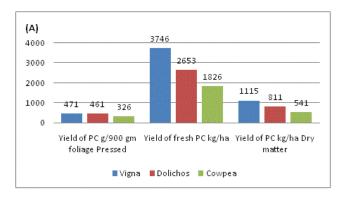


Figure 1B: The yields of green fodder, dry matter (in kg/ha) from Vigna, Dolichos and Cowpea

Table 2: The yields of pressed crop (PC) residues obtained from Vigna, Cowpea and Dolichos

Crop	Yield of PC g/900 g foliage	Pressed Crop		Yields of Pressed Crop (PC) kg/ha		
	pressed	% Dry matter (DM)	N % of DM	Fresh	Dry Matter	Crude Protein
Vigna	471	29.86	2.41	3746	1115	169
Cowpea	326	30	2.18	1826	541	74
Dolichos	461	32.6	2.31	2653	811	117
Mean	419	30.82	2.3	2742	822	120
Std. deviation	80.98	1.54	0.11	963	287	47
Std. Error	46.75	0.88	0.06	556	166	27
C.V.	19.32	4.05	4.99	35.12	34.91	39.16



**Figure 2A:** The yields of pressed crop (PC) residues obtained from Vigna, *Dolichos* and Cowpea viz., foliage pressed per 900 g, fresh pressed crop and dry matter kg/hectare

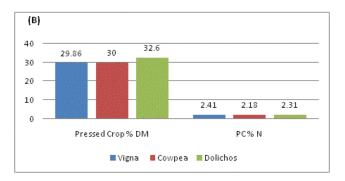


Figure 2B: The yields of pressed crop (PC) residues obtained from Vigna, Cowpea and *Dolichos* viz., percent dry matter and percent nitrogen

The yield of pressed crop/900 g of pulp was low (326 g) in cowpea, while the values for *Dolichos* and Vigna were 461 g, respectively. On the basis of the yields of green fodder, Vigna, Cowpea and *Dolichos* thus yielded 3746, 1826 and 2653 kg/hectare pressed crop residues. The PC residues in all the three crops were with high DM content and the present DM ranged from 29.8% in Vignato 32.6% in *Dolichos*. The yield of DM going in the pressed crop was 1115, 541 and 811 kg/hectare, respectively. The nitrogen content in the pressed crop residues was low than that obtained with the fresh green foliage. This was due to the removal of proteins in the juice during fractionation of the foliage. The N content of DM was 2.1% of cowpea, 2.31% in *Dolichos* and 2.41% in Vigna. The PC residue, on the basis of DM and N content, was found suitable in animal nutrition as they contained from 13 to 15% crude protein. It was also noticed that these pressed crop residues prepared good quality of silage due to its low moisture content. The yields of Crude protein going in PC residue was 74 kg/ha for Vigna. The overall results presented in Table 2 and Figures 2A and 2B give an indication that during fractionation large amount of pressed crop residue is left behind. Its immediate use in animal nutrition is desirable as it is rich in protein.

Table 3: The yields of leaf protein concentrate (LPC) prepared from Vigna, Cowpea and Dolichos

Crop	Juice released due to pressing ml/900 g green foliage	Yield of LPC g/50 ml	N % of Dry matter in LPC	Yields of LPC (kg/ha )		
				Dry Matter	Crude Protein	
Vigna	340	1.76	7.43	93	43	
Cowpea	417	1.61	8.33	71	37	
Dolichos	340	1.33	9.47	51	30	
Mean	366	1.56	8.41	72	37	
Std. Deviation	44	0.21	1.02	21	6.5	
Std. Error	25	0.12	0.58	12	3.75	
C.V.	12.02	13.46	12.12	29.16	17.56	

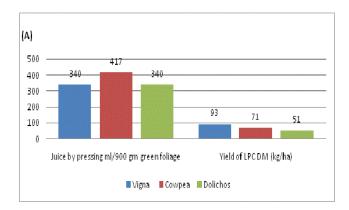


Figure 3A: The yield of juice after pressing the pulp and dry LPC in kg/ha

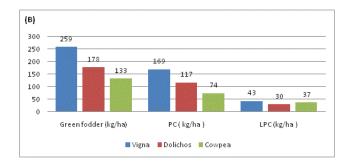


Figure 3B: Crude protein in green fodder, pressed crop and leaf protein concentrate (kg/hectare)

Other Nutrients As this product is liable for microbial deterioration and subsequent spoilage, its use in animal nutrition, as early as possible, should be made. If it is not possible to immediately use this product in the ration of animal nutrition, it has to be conserved by either drying or preparing silage.

Drying of the pressed crop residue is most simple and it can be naturally converted into a dried feed in full sunshine. Earlier work in this laboratory has shown that PC residues make good quality of silage than that made from whole unfractionated crop. Thus, it is concluded that this bulk of by-product should be used wisely in animal feeding to make the process more efficient.

Table 3 and Figures 3A and 3B indicate the amount of juice extracted per 900 g pulped foliage was 340 ml in Vigna and *Dolichos* while cowpea could extract 417 ml of juice. The amount of juice extracted showed relationship between dry matter content in the crop. Higher the dry matter, lower was a content of moisture in foliage which extracted less juice. The yield of LPC per unit volume of juice varied with species. After heating 50 ml of the juice samples *Dolichos*, Cowpea and Vigna yielded 1.33, 1.61 and 1.76 g of LPC. The yield of LPC reached 51, 71 and 93 kg/ha in *Dolichos*, Cowpea and Vigna, respectively. All the three crops yielded LPC with high nitrogen content. The N % of dry matter in LPC prepared from Vigna, Cowpea and *Dolichos* were 7.43, 8.33 and 9.47, respectively. Thus the 3 crops produced LPC containing 46 to 59% crude protein in it. The yield of LPC was 30 kg/hectare with *Dolichos*, 37 kg for cowpea, 43 kg/ha for Vigna. The overall results presented in table 3 indicated that from 30-40 kg/ha LPC could be obtained from these crops with 45 to 60% protein in it. Figure 7 indicates protein content in all green pulp or fodder, PC and LPC.

## **CONCLUSION**

There is a wide scope for increase in the yields of LPC from these crops, through the use of fertilizers, by adopting technique of intercropping, by harvesting the crop at a proper stage and by improving the method of the preparation of LPC.

#### DECLARATION OF CONFLICT OF INTEREST

The sole author declares that there is no conflict of interest.

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