

Studies on the Nutritional Evaluation of Neem Leaves of Pantnagar Area



Plant Nutrition

KEYWORDS : Nutritional, evaluation, Neem, leaves, Pantnagar

* M. K. Singh

Department of Livestock Production Management, C.V.A.Sc., G.B.P.U.A&T, Pantnagar -263 145, Uttarakhand, India * Corresponding Author

S. K. Singh

Department of Livestock Production Management, C.V.A.Sc., G.B.P.U.A&T, Pantnagar -263 145, Uttarakhand, India

S. Sathapathy

Department of Veterinary Anatomy, C.V.A.Sc., G.B.P.U.A & T, Pantnagar -263 145, Uttarakhand, India

ABSTRACT

The present work was carried out on the Neem leaves of Pantnagar area to elucidate their nutritional properties. The neem leaves of same quantity were collected from the various areas such as Department of Livestock Production Management (LPM), Instructional Poultry Farm (IPF) and Instructional Dairy Farm (IDF) inside the campus of G. B. Pant University of Agri. and Tech., Pantnagar. The leaves were identified and leaf powder was prepared to analyse for proximate analysis, i.e.; Dry matter (DM), CP (Crude Protein), CF (Crude Fibre), total ash, EE (Ether Extract), NFE (Nitrogen Free Extract), calcium and phosphorus. It was revealed that the Neem Leaf Powder (NLP) collected from the Instructional Poultry Farm had higher percentages of dry matter, CP, CF and EE, whereas the values of calcium and phosphorus were found to be more in the leaves collected from the Department of LPM of the University. It can be concluded that the NLP prepared from the leaves collected from the IPF can be included in the diet of the livestock for getting optimum results in growth performance, haematological, biochemical and immunological aspects.

Introduction

The medicinal plants and herbs have been used for many years in the treatment of various diseases in animals and human beings. Now-a-days, these are used in animal feed as the growth promoters. Due to prohibition of most of the antimicrobial feed additives in animal feed and due to their residual effects in animals, plant extracts are becoming more popular. The leaf extract of *Azadirachta indica* (Neem tree) is reported to possess diverse pharmacological activities like anti-inflammatory, hypolipidaemic, immunostimulant, hepatoprotective and hypoglycaemic effects (Khosla et al., 2000). The hypoglycemic effect was reported by various research workers (Khosla et al., 2000), where hypoglycemia was recorded in both normal and experimentally induced diabetic animals (as cited by Jaykumar et al., 2002). *Azadirachta indica* (Neem), *Zizyphus vulgaris* (Jujube berries), and *Ocimum gratissimum* (Tulsi) have the strong antibacterial activity, whereas leaves of *Azadirachta indica* are used for feeding and reducing the parasitic load of animals. The fruit of *Azadirachta indica* also has the anticoccidial activity for poultry (Tipu et al., 2006). Neem leaf and its constituents have been demonstrated to exhibit immunomodulatory, anti-inflammatory, antihyperglycaemic, anti-ulcer, antimalarial, antifungal, antibacterial, antiviral, antioxidant, antimutagenic and anticarcinogenic properties (Bhowmik et al., 2010).

Incorporation of Neem leaf Powder (NLP) in the diet of livestock had been tried by various workers, but consistent results with respect to growth performance, haematological, biochemical, immunological aspects of Neem leaf powder is lacking. Therefore, the present investigation was undertaken in view to evaluate the nutritional properties of neem leaves of Pantnagar area with an object to include them in the feed of livestock those are reared in different farms under the University.

Materials and methods

The present study was undertaken at Instructional Poultry Farm (IPF), Nagla, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, U.S. Nagar from September 2012 to December 2012. The place is located between 28° 53' 23" to 30° 27' 50" N and 77° 34' 27" to 81° 02' 22" E at 243.84 m MSL in tarai region of Uttarakhand State (India).

The Neem leaves samples were collected from the Department of Livestock Production Management (LPM), Instructional Poul-

try Farm (IPF) and Instructional Dairy Farm (IDF) inside the campus of G. B. Pant University of Agri. and Tech., Pantnagar. They were shed dried / dried at 55 ± 1°C to constant weight and ground to pass through a sieve of 1mm diameter and stored in plastic containers with lid for further analysis. Deterioration of Neem leaves occurs when it is stocked without proper drying. The fresh and wet or damaged lots should never be stocked in big pyramidal heaps for more than one day. The major active principle in Neem, azadirachtin, is highly heat sensitive. Neem leaves contain eight tetranortriterpenoids, but apparently no azadirachtins. The experiments were conducted in the moisture range of 8.0 to 10.0 per cent (w.b.). The proximate analysis on dry matter basis of Neem leaf powder studied was represented in Table 1.

Analysis of NLP

The proximate analysis viz. moisture, crude protein, crude fibre, ether extract and total ash including Ca and P of feed and moisture, crude protein, ether extract and total ash content of meat were determined as per the methods of AOAC (1995).

i. Determination of Dry Matter

Fresh sample was taken in the pre-weighted Petri dish and kept in hot air oven at 100°C for 24 hrs. Dry matter was calculated as follows:

$$\text{Dry matter (\%)} = \frac{b}{a} \times 100$$

Where,

a = Fresh weight of sample (g)

b = Weight of sample after drying (g)

$$\text{Moisture (\%)} = 100 - \text{Dry matter (\%)}$$

ii. Determination of Nitrogen and Crude Protein

The protein content was determined by Kjeldahl method for the purpose 2gm of sample was taken in a digestion flask followed by addition of 3gm of digestion mixture (K_2SO_4 : $CuSO_4$ in 9:1 ratio) and 20ml of conc. sulphuric acid. The contents were then digested till a blue/green transparent liquid was obtained. The volume of digested mixture was made up to 100ml with distilled water. A 20ml aliquot of digested mixture was distilled with ex-

cess of 40% NaOH solution and liberated ammonia was collected in 20ml of 2% boric acid solution containing 2 to 3 drops of mixed indicator (10ml of 0.1% bromocresol green + 2ml of 0.1% methyl red indicator in 95% alcohol). The entrapped ammonia was titrated against 0.1N HCl. A reagent blank was similarly digested and distilled. Nitrogen content in sample was calculated as follows:

$$\%N = \frac{\text{Sample Titre} - \text{Blank Titre} \times \text{Normality of HCl} \times 14 \times \text{Volume made up}}{\text{Aliquot of digest taken} \times \text{Weight of sample taken}}$$

Nitrogen was converted to % protein by multiplying with 6.25.

iii. Determination of Ether Extract

For estimation of ether extract Soxhlet method was used. In this method 2 gm of dried and grinded sample was transferred to a thimble. Petroleum ether (B.P. 40-60°C) was used as solvent which was subsequently evaporated. After 8 hours the thimble was taken out and it was weighed after complete drying in an oven at 60°C. Percent ether extract in dried sample was calculated as follows, which was converted into wet basis by multiplying by a moisture factor.

$$\text{Ether extract (\%)} = \frac{b}{a} \times 100$$

where,

a = weight of sample

b = (wt. of oil flask after extraction) - (wt. of oil flask before extraction)

iv. Determination of Crude Fibre

The dry sample after de-fatting was transferred from thimble to spoutless beaker of one litre capacity and in beaker; 200ml of 1.25% H₂SO₄ was added. It was refluxed for 30 minutes on hot plate after the boiling started and thereafter, filtered through muslin cloth. The residue was washed 5-6 times with hot water until it became acid free. The residual material on the muslin cloth was again transferred to the respective beaker and in beaker 200 ml of 1.25% sodium hydroxide (NaOH) solution was added. It was again refluxed for 30min. after the boiling started and thereafter filtered through muslin cloth and washed with hot water for 5-6 times until it became free from alkali. Thereafter, total residue was transferred in a clean, dry silica crucible and dried in hot air oven at 100°C for 24 hrs. Then, it was cooled in desiccator and weighed. The residue was then ignited in Muffle furnace at 600°C for 2 hrs. After 12 hrs silica crucibles containing ash were removed from the furnace and transferred into desiccator, cooled and weighed again. Weight loss during ignition was recorded as the weight of crude fibre:

$$\text{Crude fibre (\%)} \text{ on DM basis} = \frac{b-c}{a} \times 100$$

where,

a = weight of sample on DM basis (gm)

b = weight of silica crucible before ignition (gm)

c = weight of silica crucible containing residue after ignition (gm)

v. Determination of Total Ash

5 gm of ground sample was taken in previously weighed silica crucible. The crucible along with sample was kept on a heater and burnt till no more smoke was given off by the charred mass of sample. Thereafter, the silica crucible containing charred mass of sample was transferred into muffle furnace with the help of metal tong and ignited at 600°C for 2 hrs. After 12 hrs, the crucible containing ash was removed from the furnace and

then transferred into desiccator, cooled and weighed. Total ash was calculated as follows:

$$\text{Total ash (\%)} \text{ on DM basis} = \frac{a-b}{c} \times 100$$

where,

a = weight of silica crucible with ash (gm)

b = weight of empty silica crucible (gm)

c = weight of sample taken for ashing on dry matter basis (gm)

vi. Estimation of Calcium

For calcium estimation 10 ml of hydrochloric acid extract was taken in a 100 ml beaker and 2-3 drops of methyl red indicator was added to it.

Extract was heated to boiling, cooled and then 10 ml of saturated ammonium oxalate solution was added slowly with constant stirring until the precipitate became coarsely granular.

The contents were heated to boiling, cooled and then ammonium hydroxide (1: 4) was added till the colour became faint pink. The beaker was then kept overnight to settle the precipitate. The contents of beaker were then filtered through Whatman's filter paper No. 40 leaving as much precipitate as possible in the beaker, the precipitates were then washed with hot water till they became free from soluble oxalates. The point of the filter paper was then broken with a glass rod and the precipitate were then washed into the beaker in which calcium was precipitated and dissolved in about 10 ml of dilute sulphuric acid (1: 9). The contents of beaker were heated to about 60°C and titrated against N/10 KMnO₄. Finally, the filter paper was also kept in the beaker and the titration was completed. (Faint pink colour persisting for at least 30 seconds was indicative of complete titration).

Calcium (%) was calculated as under

1 ml of N/10 KMnO₄ = 0.002 g calcium

$$\text{Calcium (\%)} = \frac{\text{ml of N/10 KMnO}_4 \times 0.002 \times \text{volume of HCl extract made}}{\text{wt. of sample taken for ashing} \times \text{aliquot taken}} \times 100$$

vii. Estimation of Phosphorus

For phosphorus estimation 10 ml aliquot of HCl extract was taken into a 50 ml volumetric flask and to it 5 ml of 6.6% ammonium molybdate reagent was added. Water was then added to make the volume to 40 ml. 5 ml of 7.5 N H₂SO₄ was then added and mixed. After this 4 ml ferrous sulphate reagent was mixed and volume was made up to the mark. Optical density was read immediately in spectrophotometer at 660 nm.

The data obtained were analysed statistically as per the standard methods given by Snedecor and Cochran (1994).

Results and Discussion

The Dry matter (%), Crude Protein (%), Ether Extract (%), Total Ash (%), Crude Fibre (%), Calcium (g) and Phosphorus total (g) of NLP of leaves collected from the Department of LPM were found to be 88.24±0.03, 21.20±0.02, 2.37±0.01, 7.98±0.02, 7.81±0.04, 1.88±0.02 and 0.25±0.02 respectively (Table 1). The DM (%), CP (%), EE (%), Total Ash (%), CF (%), Calcium (g) and Phosphorus total (g) of NLP of leaves collected from the IPF were found to be 90.68±0.05, 23.40±0.01, 3.36±0.03, 8.02±0.03, 7.43±0.02, 1.40±0.01 and 0.21±0.01 respectively (Table 1). Similarly, the DM (%), CP (%), EE (%), Total Ash (%), CF (%), Calcium (g) and Phosphorus total (g) of NLP of leaves collected from the IDF were found to be 87.23±0.07, 17.50±0.01, 2.48±0.01, 9.90±0.06, 7.16±0.05 and 1.15±0.01 respectively.

From the present study, it was quite obvious that the NLP collected from the Instructional Poultry Farm had higher percentages of dry matter, CP, CF and EE, whereas the values of calcium and phosphorus were found to be more in the leaves collected from the Department of LPM of the University. There were no significant differences with respect to the values of total ash and crude fibre contents of different NLPs.

Escalating feed costs and feed ingredient availability is some of the most limiting factors affecting successful commercial livestock production in the Asian countries. This is attributable to a growing competition between human and livestock for the grains and plant proteins for which suitable alternatives need to be identified (Ravindran, 1995). Feed is an important and critical input for the livestock industry as it accounts for 60 to 70% of production cost. Commercial livestock require nutritionally balanced diet. Various feed additives or growth promoters have been developed to improve growth rate, feed efficiency and product quality and to reduce the production cost. It was quite obvious that the NLP prepared from the leaves collected from the IPF can be included in the diet of the livestock for getting optimum results in growth performance, haematological, biochemical and immunological aspects.

Acknowledgement

Authors are thankful to the Director, Experiment Station, G.B. Pant University of Agriculture and Technology, Pantnagar and Dean, College of Veterinary and Animal Sciences, Pantnagar for providing necessary facilities to conduct the experiment. Assistance provided by Department of Livestock Products Technology of G. B. Pant University of Agricultural and Technology for this research work is also thankfully acknowledged.

Conclusion

It can be concluded from the present study that the NLP prepared from the leaves collected from the IPF can be included in the diet of the livestock for getting optimum results in growth performance, haematological, biochemical and immunological aspects.

Table 1. Nutrient composition of Neem leaf powder (NLP) collected from the various areas of G. B. Pant University of Agri. and Tech., Pantnagar

Contents	Department of LPM	Instructional Poultry Farm	Instructional Dairy Farm
Dry matter (%)	88.24±0.03	90.68±0.05	87.23±0.07
Crude Protein (%)	21.20±0.02	23.40±0.01	17.50±0.01
Ether Extract (%)	2.37±0.01	3.36±0.03	2.48±0.01
Total Ash (%)	9.87±0.02	9.84±0.03	9.90±0.06
Crude Fibre (%)	7.81±0.04	7.79±0.02	7.83±0.05
Calcium (g)	1.88±0.02	1.40±0.01	1.15±0.01
Phosphorus total (g)	0.25±0.02	0.21±0.01	0.19±0.02

REFERENCE

- AOAC. 1995. Animal feeds. Official methods of analysis of AOAC International, 16th Ed. AOAC International, Virginia, USA. Vol. 1: 2201-301, pp: 1-18. | Bhowmik, D.; Yadav, C. J.; Tripathi, K. K. and Sampath, K. P. 2010. Herbal Remedies of *Azadirachta indica* and its Medicinal Application. J. Chem. Pharm. Res. 2(1): 62-72. | Jayakumar, K.; Srinivasan, M. R.; Ramesh, N.; Sachan, A.; Umesh, M. H.; Honnegowda and Narayana, K. 2002. Effect of Neem leaf extract on feed intake and body weights in rats. Indian Vet. J., 79 (7):735 -736. | Khosla, P. Bhanwara S, Sing, J, Sethi, S and Srivastava, RK (2000). A study of hypoglycaemic effects of *Azadirachta indica* (Neem) in normal and alloxan diabetic rabbits. Indian J. Physiol. Pharmacol.44:69-74. | Ravindran, V. (1965). Evaluation of layer diet formulated from non-conventional feeding stuffs. British Poultry Science. 36: 165-170. | Snedecor, G. W. and Cochran, W. B. 1994. Statistical methods. 8th Ed. The Iowa State University Press, Ames, IOWA, USA. | Tipu, M. A.; Akhtar, M. S.; Anjum, M. I. and Raja, M. L. 2006. New dimension of medicinal plants as animal feed. Pakistan Vet. J. 26 (3): 144-148. |