Full Length Article

Evaluation of nutrient composition and phenolic compound of pomegranate fruit waste as a novel feed for dairy cattle

Patil Saroja Popat[1], A.Bharathidhasan, R. Karunakaran, K. Vijayarani and P. Tensingh Gnanaraj

Department of Animal Nutrition, Madras Veterinary College, Chennai, Tamil Nadu, INDIA – 600007
Tamil Nadu Veterinary and Animal Sciences University

ABSTRACT

The pomegranate (*Punica granatum*) belongs to the *Punicaceae* family and is a nutrient dense food source rich in phytochemical compounds.About 50 % of the total fruit weight corresponds to the peel, which is an important source of bio active compounds such as phenolics, flavonoids, ellagitannins (ETs) and proanthocyanidin compounds which can be used as an alternative feed source for dairy cattle. Hence, the present study was carried out to evaluate the nutrient composition and polyphenolic compound of pomegranate fruit waste (PFW) for dairy cattle. Six samples of PFW were collected from six different places in India and screened for their nutritional composition and polyphenolic compound. The results showed that pomegranate fruit waste contained 12.72 % DM, 4.80 % CP, 12.26 % CF, 3.43 % TA, 1.47 % EE, 65.3 % NFE, 28.21 % NDF, 23.30% ADF, 4.91 % Hemicellulose, 13.79 % Cellulose and 5.14 % Lignin. Polyphenolic compound of PFW were 2.57 % total phenolic compound, 2.43 % total tannin content,0.13 % non-tannin phenolics, 1.75 % condensed tannin, 0.69 % hydrolysable tannin and 2.05% total flavonoid content.Thus, it is concluded that PFW is of high nutritive value which can be used for feeding to dairy cattle.

Key words : Pomegranate fruit waste, Nutrient composition, Polyphenolic compound.

INTRODUCTION

The pomegranate (*Punica granatum*) belongs to the *Punicaceae* family and is a nutrient dense food source rich in phytochemical compounds. Pomegranate fruit waste is a by-product of the pomegranate juice industry. PFW can be used as a relatively good agro-industrial by-product for ruminant nutrition (Mirzaei Aghsaghali *et al.,* 2011). Pomegranate fruit consists of seeds, aerials and peel which include the husk and interior network membranes. Pomegranate Fruit Waste (PFW) contains protein, carbohydrates, lipids, vitamins, minerals and water. India is one of the largest producers of pomegranate in the world. The annual production of pomegranate is about 13.46 lakh tonnes in India (NRCP, 2015). About 50 % of the total

[1] Corresponding author:
Email: sarojapatil9233@gmail.com

fruit weight corresponds to the peel, which is an important source of bio active compounds such as phenolics, flavonoids, ellagitannins (ETs) and proanthocyanidin compounds. Climatic condition and water scarcity had escalated the cost of animal feeds. Use of agricultural by-products obtained after processing of fruits such as pomegranate pulp by-product is one of the useful way of overcoming this problem. In the last few years there is an increasing interest of nutritionists in bio-active plant factors like phytochemicals as natural feed additives. Tannins are the ones that can modify the rumen fermentation processes, improve the protein metabolism, reduce ammonia production and curb methane emission to the atmosphere. In a livestock production system, pomegranate fruit waste is found to be appropriate for use in animal feeds due to the presence of various micronutrients. The addition of pomegranate fruit waste extraction using either water or solvent mixture positively altered some rumen parameters such as NH_3–N concentration, acetate-to-propionate ratio and protozoan growth; increased microbial protein and also improved the yield and quality of milk (Abarghuei *et al*., 2013). Hence, the present study was carried out to evaluate the nutrient composition and phenolic compound of pomegranate fruit waste for feeding dairy cattle.

MATERIALS AND METHODS

Six samples of PFW were collected from six different places in India viz. Chennai, Solapur, Sangli, Kancheepuram, Thanjavur and Tiruchirapalli. The PFW sample were dried in hot air oven at a temperature of 55-65°C to constant weight and ground to pass through 1mm sieve and stored in airtight containers for further analysis. Dry matter (DM) was determined by drying the samples at 105°C overnight and ash by igniting the samples in muffle furnace at 525°C for 8 hours and Nitrogen (N) content was measured by the Kjeldahl method (AOAC, 1990). Crude fiber is determined by acid digestion and alkali digestion of pomegranate fruit waste. Crude protein (CP) was calculated as N × 6.25. Moisture, crude protein, ether extract, total ash (TA) and crude fiber (CF) contents of pomegranate fruit waste were determined according to the procedures of the AOAC (2000). The fibre fraction of pomegranate fruit waste was determined according to procedure of Van Soestand Robertson(1976).

For estimation of polyphenolic compound, the PFW extracts were prepared and estimated as per the method explained by Makkar *et al*. (1993).The measurement of total phenolic (TPs) content was conducted according to the modified Folin–Ciocalteu colorimetric method. Total phenolics were estimated by adding 0.1 ml of PFW extract with the following *viz.* 0.9 ml of distilled water, 0.5 ml of 1N Folin Ciocalteu reagent and 2.5 ml of 20 % sodium carbonate solution. The contents were mixed and incubated for 40 minutes at room temperature. The optical density was measured at 725 nm in a UV spectrophotometer. The concentration was calculated with tannic acid equivalent standard curve.The standard curve was drawn from different concentrations of standard tannic acid solution (0.1mg/ml)

ranging from 0 to 0.3 ml by adopting the same procedure as that for plant extracts.

For non-tannin phenolics estimation, 100 mg of poly vinyl poly pyrrolidone was taken in test tube and to it one ml of distilled water along with 1 ml of PFW extract was added. Then it was centrifuged at 3000 rpm for 10 minutes and the supernatant was collected. The supernatant contained non tannin phenolics. Total tannin phenolics were calculated by subtracting non tannin phenolics from the total phenolics.

The condensed tannin present in the PFW extracts were estimated as per the method of Porter *et al*. (1986) where in 0.5 ml of PFW extract was taken in test tube and to it was added 3ml of butanolHCl and 0.1 ml of ferric reagent. The test tubes were vortex mixed to ensure proper mixing. The mouth of the tubes was covered with glass marble and contents boiled for 60 minutes. The tubes were cooled to room temperature and optical density was read at 550 nm using spectrophotometer. Condensed tannin as leucocyanidine equivalent was calculated by using the following formula.

$$\text{Percentage of condensed tannin} = \frac{\text{Absorbance at 550 nm x 78.26 x dilution factor}}{\text{Percentage of dry matter}}$$

The hydrolysable tannin was calculated by subtracting condensed tannins from total tannins (Singh *et al*., 2005).

Total flavonoid content was measured by the Aluminum chloride ($AlCl_3$) colorimetric assay (Jia *et al*., 1999). An aliquot of 1 ml PFW extract was added to 10 ml volumetric flask containing 4 ml of double distilled water. Then 0.3 ml of 5 % $NaNO_2$was added to the flask and after 5 minutes, 0.3 ml $AlCl_3$(10 %) was also added. At 6^{th} minute, 2 ml NaOH (1 M) was added and the total volume was made up to 10 ml with double distilled water. The solution was mixed completely and the absorbance level was measured at 510 nm against reagent blank. Six different concentrations of quercetin solutions (20–100 mg/l) were prepared and used for calibration of standard curve.All data were statistically analysed as per the Snedecor and Cochran (1989).

RESULTS AND DISCUSSION

The results of proximate composition of PFW showed that 12.72% DM, 4.80 % CP, 12.26 % CF, 3.43% TA, 1.47 % EE, 65.3 % NFE, 28.21 % NDF, 23.30 % ADF, 4.91 % Hemicellulose,13.79 % Cellulose and 5.14 % Lignin. These results were in conformity to the findings of Mirzaei-Aghsaghali*et al.*(2011),who reported the DM, CP, EE, NDF, ADF and NFC content of pomegranate were 13.01, 3.60, 0.61, 20.80, 15.10 and 69.57 %, respectively. Further the present result of chemical composition was consistent with earlier report by Kushwaha*et al.* (2013) and Rowayshed*et al.*(2013). The proximate composition of PFW did not vary much with that analysed in present study as well as earlier reports. Hence pomegranate fruit waste can be used as an alternative resource for ruminants.

Further the polyphenolic content of PFW were 2.57 % total phenolic compound, 2.43 % total tannin content, 0.13 % non-tannin phenolics, 1.75 % condensed tannin, 0.69 % hydrolysable tannin and 2.05 % total flavonoid content.The present findings of polyphenolic compound of PFW were almost consistent with earlier reports by Yunfeng and Changjiang (2005). Also the total phenolic compound of PFW were also similar with those found by Kushwaha*et al.* (2013). However,Besharati (2015) reported the total phenolic (1.8 %) and total tannin (0.8 %) content of pomegranate waste which were considerables lower than the present study.

In a livestock production system, pomegranate fruit waste found to be appropriate for use in animal feeds due to the presence of various micronutrients. Addition of supplements such as ionophores, antibiotics, defaunating agents and methane inhibitors to ruminants improve their fermentation, which subsequently improves the milk and meat efficiency (Abarghuei *et al*., 2013). Dkhil (2013) demonstrated that the methanolic extract of Pomegranate peel (PP) has natural antioxidant capacity and it also has anthelmintic activity. Because of its high nutritional value and antioxidant capacity, PP is a health-promoting food source for animals, especially ruminants. Shabtay *et al.* (2008) demonstrated that dietary supplementation of pomegranate peels promoted increase in feed intake with a tendency to increase weight gain in bull calves.

Ruminants, have the unique capacity to utilize fibre, because of the presence of rumen microbes. This means that cereals can largely be replaced by pomegranate by-products (Mirzaei-Aghsaghali*et al*., 2011). Shabtay*et al.* (2008) concluded that different pomegranate components may have different nutritive effects and influence milk production in different ways. Therefore, based on present study it is concluded that pomegranate fruit waste is nutrient rich feed source which can be used for feeding to dairy cattle to increase the production and performance of animal.

REFERENCES

Abarghuei, M.J., Rouzbehan, Y., Salem, A.Z.M and Zamiri, M.J.(2013). Nutrient digestion, ruminal fermentation and performance of dairy cows fed pomegranate peel extract. *Livest. Sci*., 157; 452-461.

AOAC. (1990). Official Methods of Analysis. 15th Edn. Washington DC. USA, Association of Official Analytical Chemists. pp. 66-88.

AOAC. (2000). Association of Official Analytical Chemists. Official Methods of Analysis of the AOAC International, 17th ed. Gaithersburg, Maryland, USA.

Besharati, M. (2015). Evaluation of Saccharomyces cerevisiae supplemented pomegranate seed using in vitro gas production technique. *Slovak J. Anim. Sci*., 48, 2015 (**2):** 64–70

Dkhil, M.A., (2013). Anti-coccidial, anthelmintic and antioxidant activities of pomegranate (Punica granatum)

peel extract. *Parasitol. Res.* 112, 2639–2646.

Jia Z, Tang M and Wu J., (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chem*.**64**: 555-559.

Kushwaha, S. C., Bera, M. B. and Pradyuman Kumar.(2013). Nutritional Composition of Detanninated and Fresh Pomegranate Peel Powder. I*OSR Journal of Environmental Science, Toxicology and Food Technology* 7, 1(**7**): 38-42.

Makker, H.P.S, M. Blummel, N.K.Borowy and K. Becker. (1993). Gravimetric determination of tannins and their correlations with chemical and protein precipitation methods. *J.Sci. Food. Agric.,* **61**: 161-165.

Mirzaei-Aghsaghali. A., Maheri-Sis, A. N., Mansouri, H., Razeghi, M.E., Cheraghi, H. and Aghajanzadeh-Golshani, A. (2011).Evaluating potential nutritive value of pomegranate processing by-products for ruminants using *in vitro* gas production technique. *ARPN J. Agric. Biol. Sci*. **6:** 45-51.

NRCP, (2015). National Research Center on Pomegranate, Solapur. *Vision 2050.* pp. 1.

Porter, L.J., Hrstich, L.N. and Chan, B.J. (1986). The conversion of proanthocyanidines and prodelphinidins to cyaniding and delphinidin. *Phytochemistry.,* **25**: 223-230.

Rowayshed G. and Salama, A., (2013). Nutritional and Chemical Evaluation of Pomegranate (Punica granatum L.) Fruit Peel and Seeds Powders by Products. *Middle East Journal of Applied Sciences,* 3(**4**): 169-179.

Shabtay, A., Eitam, H., Tadmor, Y., Orlov, A., Meir, A., Weinberg, P., Weinberg, Z.G., Chen, Y., Brosh, A., Izhaki, I., Kerem, Z. (2008). Nutritive and antioxidative potential of fresh and stored pomegranate industrial byproduct as a novel beef cattle feed. *J. Agric. Food Chem*. **56:**10063–10070.

Singh, B., A. Sahoo, R. Sharma and T.K. Bhat, 2005. Effect of polyethylene glycol on gas parameters and nitrogen disappearance of some tree forages. *Anim. Feed Sci. Technol.,* **123-124**: 351-364.

Snedecor, G.W. and W.C. Cochran, (1989). Statistical Methods, 8thedn., Iowa State University Press, Ames, Iowa.

Van Soest, P.J. and J.B. Robertson, (1976). Chemical and physical properties of dietary fibre. In Dietary Fibre; Proceedings of the Miles Symposium.

Yunfeng, L., Changjiang, G. (2005). Evaluation of antioxidative properties of pomegranate peel extracts in comparison with pomegranate pulp extract. *Food Chemistry* **96**: 254-260.