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Amla as phytogenic feed additive for efficient livestock production

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Abstract
Indian gooseberry or amla (*Phyllanthus emblica Linn or Emblica officinalis Gaertn*.) is a valuably most important medicinal plant in Indian traditional system of medicine, *i.e*. in Ayurveda. Various parts of the plant are used to treat a range of diseases or used as feed additives in animals but the most important part is fruit. The fruit is rich in quercetin, phyllaemblic compounds, gallic acid, tannins, flavonoids, pectin and vitamin C and also contains various polyphenolic compounds. A wide range of phytochemical components including terpenoids, alkaloids, flavonoids and tannins have been shown to posses useful biological activities. The fruit is used either alone or in combination with other plants and have wide range of action such as antipyretic, analgesic, antitussive, antiatherogenic, adaptogenic, cardioprotective, gastroprotective, antianemia, anti hypercholesterolemia, wound healing, antidiarrheal, antiatherosclerotic, hepatoprotective, nephroprotective and neuroprotective properties. Amla is also reported to possess radiomodulatory, chemomodulatory, chemopreventive effects, free radical scavenging, antioxidant, anti-inflammatory, antimutagenic and immunomodulatory activities. In this review, we have focused our interest on use of amla as phytogenic feed additive for efficient livestock production.

Keywords: amla, feed additive, livestock, medicinal plant

Introduction
Amla has been regarded as the sacred tree in India. The tree was worshipped as Mother Earth and is believed to nurture mankind because the fruits are very nourishing. The leaves and fruits are used in worship in India. According to Indian mythology it is believed to be the first tree created in the universe. The medicinal properties of amla have been mentioned in the old Ayurvedic text such as Charaksanhita and Sushrutsanhita. Amla is a monoecious glabrous or pubescent deciduous tree belongs to the family Euphorbiaceae. The name of the family Euphorbiaceae comes from Euphorbus, physician to Juba, King of Mauritania. The plant is commonly known as Amla and found throughout plains of India, Pakistan, Nepal, Uzbekistan, Sri Lanka, SE Asia, China and Malaysia at an elevation of 610-1390 m above the sea level [1, 2]. The Persian name of Emblica is amlah (from Sanskrit amla) but, as the Arabic amlaj suggests, probably in older Persian amlag, and hence Emblica. Garcia de Orta says it was called embelgi by the Arab physicians. In Sanskrit amla has many synonym: Dhatriphala (dhatri means female supporter, a nurse; phala means fruit); Vayahstha (means strong, vigorous, 'being in the bloom of age'); Amritaphala (amrita means immortal); Amala (means pure or clean); Amla (sour) all these synonymous words show how important this plant is in traditional Indian medicine, in fact it is the most common Ayurvedic plant [3].
The Amla fruits are sour, astringent, bitter, acrid, sweet, cooling and anodyne in nature and used as ophthalmic, carminative, digestive, stomachic, laxative, alterant, aphrodisiac, rejuvenative, diuretic and antipyretic purposes. They are useful in vitiated conditions of tridosha, diabetes, cough, asthma, bronchitis, cephalalgia, ophthalmopathy, dyspepsia, colic, flatulence, hyperacidity, peptic ulcer, erysipelas, skin diseases, leprosy, haematogenesis, inflammations, anemia, emaciation, hepatopathy, jaundice, strangury, diarrhoea, dysentery, hemorrhages, leucorrhoea, menorrhagia, cardiac disorders, intermittent fevers and greyness of hair.

Taxonomy
Kingdom: Plantae
Division: Angiosperm
Class: Magnoliopsida
Order: Euphorbiales
Family: Euphorbiaceae
Genus: *Phyllanthus*
Species: *emblica L*

Because of all these properties amla is explored as herbal feed additive to enhance the livestock and poultry production. Very less work is reported in case of ruminant animals as compared to monogastric animals. The herbal feed supplement (feed additive) is a substance or mixture used in minor quantity other than basic feed in order to complement certain nutrients for improving performance of the animals and birds. The efficiency of poor quality roughages utilization can be maximized by the supply of deficient nutrients like nitrogen, micronutrients, feed additives *etc*. in the ration of animals. Recently, growth promoters have been banned in animal nutrition. With the ban on use of antibiotics as growth promoter by European Union from March 2006, it has become need of the hour to produce drug/chemical free products. There are two principal reasons behind the changes in legislation on the use of in-feed antibiotic growth promoters. The first is to try to combat the development of microbial resistance to antibiotic drugs and its consequences on human health. The second is a response to consumer pressures to eliminate the use of all non-plant xenobiotic agents from the animal diets but at the same time, we can't afford to compromise with the production level because of increasing demand of animal products due to increasing human population and awareness of consumer regarding the importance of animal protein. Reviews on the usage pattern of phytochemicals reveals that tannins, saponins, essential oils, anthraquinones, flavonoids are the major plant secondary metabolites studied in ruminants [4, 5]. These phytochemicals could act alone or a combination of the following potential mechanisms on animal system: (i) increased feed intake (ii) improved feed efficiency (iii) improved gut function leading to reduced incidence of digestive disturbances (iv) stimulation of digestion (v) methane inhibition (vi) immune modulation (vii) antioxidant and (viii) antimicrobial effect. Usually mixture of several herbs rather than single one is more effective for better and sustainable performance of animals.

Nutritional composition of amla

Amla is well known for its nutritional qualities. It is rich in polyphenols, minerals and is regarded as one of the richest source of vitamin C (200-900mg/100g of edible portion), therapeutic potential of the fruits was attributed to their high content of ascorbic acid [6]. Amla is often incorporated in the herbal formulation called "Triphala", which contain the equal proportion of the fruits of *E. officinalis*, *Terminalia chebula* and *Terminalia belerica*. Amla fruits contain moisture 81.2%, protein 0.5%, fat 0.1%, mineral matter 0.7%, fiber 3.4%, carbohydrates 14.1%, Ca (0.05%), K (0.02%), Fe (1.2 mg/100g), nicotinic acid (0.2 mg/100g), vitamin C (600mg/100g) [7]. Amla fruit powder contains 4.23% protein, 0.5% fat, 2.38% ash, 0.32% AIA, 27.94% NDF, 19.32% ADF, 8.62% HC, 92.87% T-CHO, 0.07% Ca and 0.008% P [8]. Its ascorbic acid level ranges from 3.0 to 5.43 g/100 g of dried powder [9].

Chemistry of amla

The active ingredient that has significant pharmacological action in amla is designated by Indian scientist as "Phyllemblin". The fruit is rich in quercetin, phyllaemblic compounds, gallic acid, tannins, flavonoids, pectin and vitamin C and also contains various polyphenolic compounds. A wide range of phytochemical components including terpenoids, alkaloids, flavonoids and tannins have been shown to posse's useful biological action [10]. The fruits, leaves and bark are rich in tannins. The root contains ellagic acid and lupeol and bark contains leucodelphinidin. The seeds yield a fixed oil (16%) which is brownish-yellow in colour. It contains linolenic acid (8.8%), linoleic acid (44.0%), oleic acid (28.4%), stearic acid (2.15%), palmitic acid (3.0%) and myristic acid (1.0%) [11]. The phytochemicals of this plant include hydrolysable tannins (Emblicanin A, Emblicanin B, punigluconin, pedunculagin), flavonoids, alkaloids (Phyllantidine and phyllantine) [12]. Gallic acid, ellagic acid, chebulinic acid, quercetin, chebulagic acid, corilagin together with isostrictinnin, were isolated from the fruit of Phyllanthus emblica [13]. It contains two new hydrolysable tannins with low molecular weight (<1000), called emblicanin A (37%), emblicanin B (33%), punigluconin (12%) and pedunculagin (14%) [12]. These two new tannins (emblicanins A and B) have a very strong antioxidant action [14, 15] and found to preserve erythrocytes against oxidative stress induced by asbestos, generator of superoxide radical.

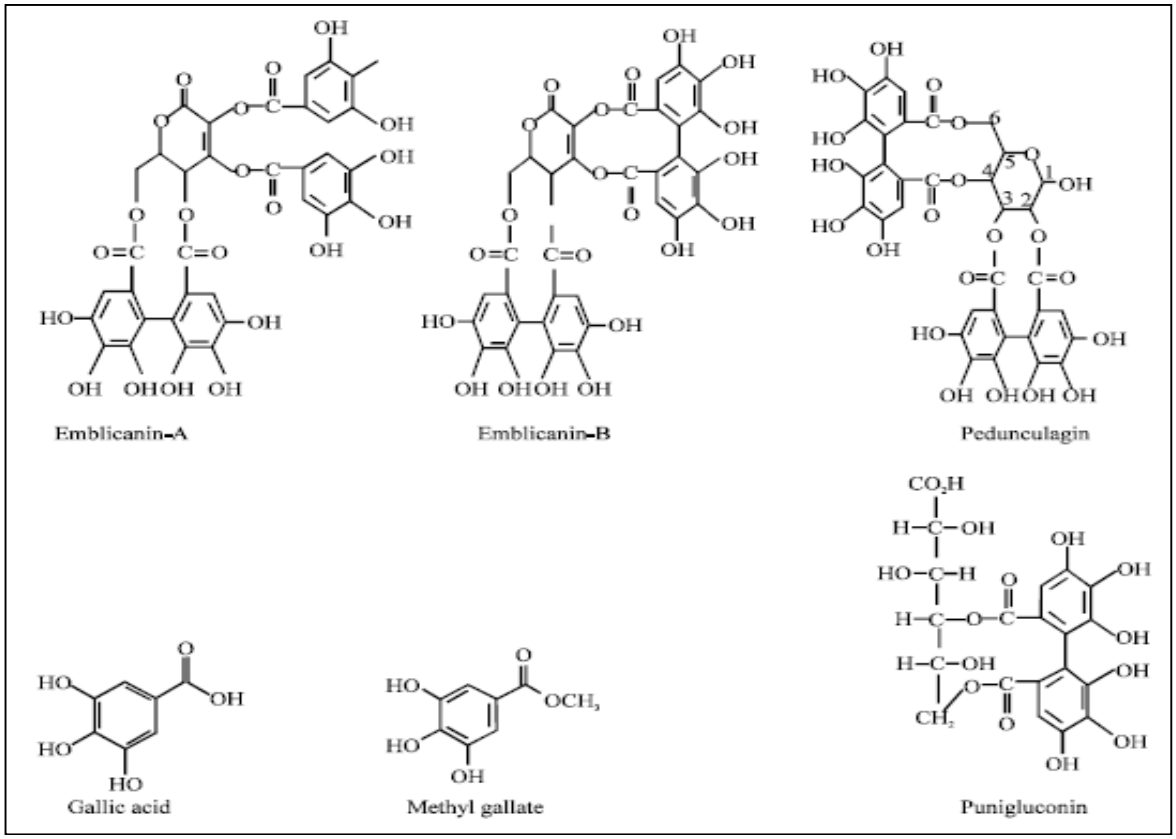

Fig: Important phytochemicals of amla

Tannins traditionally been considered as anti-nutritional factors but now a day they are more known in ruminant nutrition for their beneficial effects on ruminal modification [16]. They are classified into hydrolysable and condensed tannins and are considered to have both adverse and beneficial effects depending on their concentration and nature besides other factors such as animal species, physiological state of the animal and composition of the diet [17]. Feeding of tannin rich diet also induced a shift in rumen microbial profile with increased population of tannin tolerant bacteria [18]. The whole concept of farming during the last two decades or so has undergone a sea of changes. The poultry industry has now emerged as a highly structured and market-oriented enterprise. The major objective of livestock and poultry farming is to increase the profit margin in business by improving feed efficiency and growth rate keeping the health status of animals.

Preparation of the amla powder

Sorting and grading is essential to get suitable quality of fruit which was done by hand. The fruits were first washed to remove the dirt. Grading of fruit was done based on soundness, firmness, cleanliness, size, maturity, weight, color, shape and freedom from foreign matters, insect damage and mechanical injury. From the graded amla the pulp was extracted manually. It was homogenized in a mixer to obtain fine pulp or dry in oven at 60 ˚C then grind to obtained amla powder.

Effects on digestion

The Amla fruits have well known action as carminative, digestive, stomachic, laxative and tonic in nature. The regular use of Amla can strengthen digestion, absorption and assimilation of food. But it works more slowly and gently than ginger or other digestion-enhancing herbs. In addition, it improves assimilation of iron for healthy blood. Amla purify the Rasa Dhatu (nutrient fluid) and Rakta Dhatu (blood), thus supporting the functions of the liver. It also strengthens the liver and helps in detoxification and removal of toxins from the body [19].

Amla as feed additives have no side effects on the health of birds. The dietary use of herbal growth promoter increases the performance of broiler by improving digestion, metabolism and absorption of certain nutrients which ultimately improve live weight gain and FCR [20]. Herbs have a complex mode of action on various organs or systems due to active ingredients present in them. They are supposed to have greater impact on different factors which promote growth and health, by improving physiological and immunological functions of body. Manoj Kumar and Singh (2005) [21] conducted an experiment on day old broiler chicken of Hubbard strain to evaluate the efficacy of amla fruit on the productive performance of broiler chicken. The production was comparatively higher in amla fed group as evidence by higher body weight gain and FCR. The mortality percentage was zero in treatment groups whereas 5% mortality was recorded in control group. Likewise, Daisy *et al.* (2007) [22] supplemented amla fruit powder in water and observed growth performance of broiler. The result showed better growth and feed utilization in treated groups as compared to control.

Wadhwa *et al.* (2007) [23] observed the effect of amla fruit powder in feed of broiler at level of 250 (AM_1), 500 (AM_2) and 750 (AM_3) gm/ton and compared it with zinc bacitracin supplemented and non-supplemented groups. The body weight was significantly high in amla treated group than zinc bacitracin treated birds but there was no significant difference from weight of broiler reared without antibiotic. Feed consumption was high in birds supplemented with amla powder. During the last two weeks of age, however, the broilers in AM_3 gain significantly highest weight than rest of treatments. Singh *et al.* (2008) [24] conducted an experiment

under hot and humid climate to study the effect of density, protein and amla levels on performance of broilers. Body weight was significantly influenced by density at earlier (2 to 3 wk) and later ages (5 and 6 wk) during hot humid climate. At earlier age D_2 medium density was optimum whereas in later of age sixth week body weight was significantly high in D_3 low density than D_1 high and D_2 medium density groups. Effect of protein was significant on weight gain. Amla additive improved body weight by 14 gms at 6 weeks of age. D_3 low density has significantly better feed efficiency than D_1 high density. Tiwari *et al*. (2008) [25] studied the effect of probiotic and herbal supplement on growth and immuno competence traits of commercial broilers. The bird of the first group were fed on a basal diet supplemented with probiotic (Biovet YC @ 50 g/quintal of the basal diet) while the second group birds were fed on a basal diet supplemented with herbal mixture (1% amla pulp powder, 0.5% turmeric powder, 0.5% neem leaf powder). The herbal supplemented group had apparently higher body weight gain compared to the control group throughout the experiment and at 6th week of age the average weekly body weight gain was significantly higher in the herbal supplemented group compared to the probiotic and control groups. No significant differences in the weekly FCR of commercial broilers among different treatment groups were observed during the experimental period except at 6th week where herbal supplemented group had significantly better FCR compared to the probiotic and control group. HA titre was significantly higher in the herbal supplemented group compared to the probiotic and control group at 6 weeks of age.

Ghavate *et al*. (2009) [26] conducted an experiment to study the effect of feeding different levels of amla on performance of broilers during 0-6 weeks of age. There was significant effect of supplementation on gain in body weight of broilers in T_2 group as compared to those in T_1 and T_0 (control) groups. Feed intake was also significantly higher in T_2 group than in T_1 and control T_0 groups. The supplementation of amla powder @ 15 kg/t of feed T_2 group significantly improved FCR compared to T_1 group (2.66). It was concluded that the effect of addition of amla powder on live weight gain, feed intake and feed conversion ratio was significant due to supplementation of amla powder @ 10 kg/t and 15 kg/t of feed but costly as compared to control group. Similarly, Ghule *et al*. (2010) [27] conducted a comparative study of amla and vitamin C on performance of broilers under nutritional stress for a period of 42 days, and found that birds fed low energy (5% than control) rations with supplements amla and vit. C showed apparently higher body weight gain in fourth and sixth week than control and low energy group without supplementation. Patel *et al*. (2016) [28] also reported similar result and concluded that, dietary addition of amla fruit powder (@ 0.4 and 0.8%) had a positive effect on growth performance and net profit per bird in commercial broiler chickens as compared to control group. Similarly, Tangade (2007) [29] studied the effect of supplementation of herbal amla and synthetic vitamin C on performance of broilers during intense summer conditions. Control group T_0 received corn soya based standard ration. Group T_1, T_2 and T_3 received corn soya based standard ration supplemented with 150, 200 and 250 mg/kg synthetic vitamin C respectively. Group T_4, T_5 and T_6 received corn soya based standard ration supplemented with 150, 200 and 250 mg/kg vitamin C equivalent to synthetic vitamin C through amla. He find that the group T_5 (200 mg/kg herbal (amla) vitamin C) showed significant improvement over all groups in body weight, weekly gain in body weight, feed consumption and FCR.

Other than health promoting function of amla, some phytogenic feed additives have also shown the effect on the rumen ecosystem of ruminants, which resulted in reduced methanogenesis [30, 31] and also increased the feed utilization efficiency. The overall literature review showed that the use of amla fruit powder as herbal supplement was beneficial in livestock and poultry for its encouraging results in relation to growth performance. The overall performance and economical benefits were better due to addition of this herb to poultry rations.

Gastroprotective effects

An ethanol extract of amla was examined for its antisecretory and antiulcer activities employing different experimental models in rats, including pylorus ligation Shay rats, indomethacin, hypothermic restraint stress induced gastric ulcer and necrotizing agents (80% ethanol, 0.2 M NaOH and 25% NaCl). Oral administration of amla extract @ 250 mg/kg and 500 mg/kg significantly inhibited the development of gastric lesions in all test models used. It also caused significant decrease of the pyloric-ligation induced basal gastric secretion, titratable acidity and gastric mucosal injury. Histopathological analyses are in good agreement with pharmacological and biochemical findings. The results indicate that amla extract possesses antisecretory, antiulcer and cytoprotective properties.

Hypolipidaemic effect

The lipid lowering and antiatherosclerotic effects of amla fresh juice were evaluated in cholesterol fed rabbits (rendered hyperlipidaemic by atherogenic diet and cholesterol feeding). Fresh juice of amla was administered @ 5 ml/kgBW/day for 60 days. Serum cholesterol, TG, phospholipid and LDL levels were lowered by 82, 66, 77 and 90%, respectively [32]. Similarly, the tissue lipid levels showed a significant reduction following amla juice administration. Aortic plaques were regressed. Amla juice treated rabbits excreted more cholesterol and phospholipids, suggesting that the mode of absorption was affected. Similarly, beneficial effects of amla fruit extract on serum and tissue lipid parameters have also been reported by others [33]. Amla fruits are a rich source of ascorbic acid (600 mg %) and acts as an antioxidant to prevent the oxidation of LDL cholesterol and thus slows atherogenesis. A substantial reduction in the risk of atherosclerosis in subjects with high plasma concentrations of vitamin C has been well established [34].

Antioxidant effect

As a result of metabolic reactions and diseases, there is generation of free radicals in the form of reactive oxygen species and reactive nitrogen species within the animal's body. Free radicals are chemical entities, which contains one or more unpaired electrons due to which they are highly unstable and cause damage to other molecules by extracting electrons from them in order to attain their stability. Reactive oxygen species (ROS) are highly reactive oxidizing agents belonging to the class of free-radicals. The free-radical initiate chain reactions by removal of an electron from another molecule to complete its own orbital. The most common ROS that have potential implications in reproductive health includes superoxide anion (O^{-}_2), hydrogen peroxide (H_2O_2), hydroxyl radical (OH^-) and nitric oxide (NO^+). An increased production of ROS by any cell beyond the capacity of the

cellular antioxidant defense leads to oxidative stress (OS), which ultimately cause significant cell damage through irreversible modification of vital cellular structures such as lipids, proteins and nucleic acids. This higher shift of oxidative stress may lead to several disorders and could be pre-disposing factor for them [35]. The natural mechanism to counteract the effect of reactive oxygen species is via antioxidant compounds (vitamin C, vitamin E, carotene, uric acid) and enzymes (catalase, superoxide dismutase, glutathione peroxidase/reductase). Herbs are known to contain large amounts of different families of antioxidants of which flavanoids and tannins are most recurring followed by phenolics, ascorbic acid and alkaloids [36].

Because of two new tannins, amla have a very strong antioxidant action. The two emblicanins A and B have been found to preserve erythrocytes against oxidative stress induced by asbestos, generator of superoxide radical. Emblicanin A oxidates when put in contact with asbestos becoming emblicanin B and together they have a stronger protective action to erythrocytes than vitamin C. Moreover they improve the efficacy of vitamin C in reducing dihydroascorbic acid to ascorbic acid. The same recycling process has been observed in the rutin- vitamin C combination. Antioxidant compounds can help to protect DNA from the damage associated with free radicals which are highly reactive compounds.

Amla and other rasayanas are effective broad spectrum antioxidants and free radical scavengers, helping to reduce disease and slow the aging process. The use of amla as an antioxidant has been examined by a number of authors [37]. Adaptogenic herbs like Amla, Ashwagandha, Tulsi and Ginseng etc. are being used as anti-stress factors for long years in human and animal medicines with proven results [38]. These compounds in herbs have the capacities to quench lipid peroxidation, prevent oxidative DNA damage, and scavenge ROS, such as superoxide, hydrogen peroxide and hydroxyl radicals [39]. The oral administration of flavonoids from amla at a dose of 10 mg/kg showed significant antioxidant action in cholesterol fed experimental rats. The activities of free radical scavenging enzymes were significantly elevated and lipid peroxide content was significantly decreased in flavonoid treated hypercholesterolemic rats [40]. This antioxidant activity of the amla extract has been found to be both dose and concentration dependent. Administration of amla (@ 500 mg/kg BW for 30 days) prevents the restrain stress induced oxidative stress and elevation of corticosterone levels [41]. Maini *et al.* (2007) [42] conducted a trail on commercial broiler chicks. Control group was given basal diet and treatment groups were supplemented with either vitamin E (200mg/kg feed), dried mint leaves (10g/kg and 30g/kg feed) or amla-electrolyte mix (1g/kg feed) from day 1 to seven weeks of age. Antioxidant supplemented groups showed significantly reduced level of lipid peroxidation in erythrocytes both at 3rd and 5th week, whereas in control group at 3rd and 5th week showed high concentration of lipid peroxidation and low activity of glutathione (GSH) and superoxide dismutase (SOD) in erythrocytes, heart, liver and brain cortex.

Effect on immune modulation and immune response

Immunity is a homeostatic process, a series of delicately balanced complex, multicellular and physiologic mechanisms that allow an individual to distinguish foreign material from "self" and neutralize and/or eliminate the foreign matter [43]. The immune system is primarily concerned with resistance against foreign invaders and protection against neoplastic cells. Whereas, immunomodulator is an agent that intensifies or diminishes the immune responses and such effect is called as immunomodulatory effect. Immunomodulation using medicinal plants could provide an alternative to conventional chemotherapy for a variety of diseases, especially when host defence mechanism has to be activated under the conditions of impaired immune response or when a selective immunosuppression is desired in situations like autoimmune disorders [44]. Amla acts as strong immunity booster, antibacterial, antifungal, antiviral medical studies conducted on amla fruit suggest that it has antiviral properties [45] and also functions as an antibacterial and anti-fungal agent [46]. Amla fruit powder as feed additive has been reported to possess antistress, adaptogenic, immunogenic and growth-stimulating properties resulting in better performance of broiler [47, 23].

Anticancer and antiulcer effects

It was tested *in vivo* in wistar rats treated with carcinogen Diethylnitrosamine (DEN) (200mg/kg b.wt i.p) to induce liver cancer. The results showed that pretreatment of methanolic fruit extract (100 and 200 mg/kg b.wt) exhibited significant pathological manifestations at both the doses. Amla has the potential to be useful in ameliorating the carcinogen-induced response in rat [48]. Similarly, potential anticancer effects of aqueous fruit extract of amla was tested in several different human cancer cell lines such as A549 (lung), HepG2 (liver), HeLa (cervical), MDA-MB-231 (breast), SKOV3 (ovarian) and SW620 (Colorectal). Amla extract significantly inhibited the growth of several human cancer cell lines at doses of 50-100 µg/ml [49]. Research found that amla is benecifical to treat different types of cancers. Garima *et al*. (2005) [50] also reported chemopreventive potential of amla against skin carcinogenesis. Similarly, Banu *et al.* (2004) [51] also experimentally evaluated ethanolic extract of amla against genotoxicity induced by DMBA. Amla fruit administered orally at different concentrations (100, 250, 500 mg/kg b.wt) for seven consecutive days in Swiss albino mice prior to a single intraperitoneal injection of DMBA decreased the frequency of bone marrow micronuclei. The protection provided by amla may be due to its antioxidant capacity and through its modulatory effect on hepatic activation and detoxifying enzymes.

Antimicrobial effects

Herbal growth promoters due to their multidirectional actions are also useful in controlling pathogenic bacteria without possibility of development of resistance and are also safe for animals and humans. In such manner they can work as herbal antimicrobial agents with other advantages like more economical, free from possibility of resistance, maintaining normal functions, improving production performance in broilers, improving egg size, lymphocyte percentage and egg weight in layers [52, 53]. Saeed *et al*. (2007) [54] also reported that, aqueous infusion and decoction of amla exhibited potent antimicrobial activity against different strains of *Staphylococcus aureus*, *S. haemolyticus*, *S. saprophyticus*, *Micrococccus varians*, *M. lylae*, *M. roseus*, *M. halobius*, *M. sedenterius*, *Bacillus subtilis*, *B. megaterium* and *Candida albicans*.

Conclusion

Research on herbal plants has gained a renewed focus recently. The prime reason is that other system of feed additives although effective with a number of side effects

along with residue in final products that often lead to serious complications in human being as well as welfare of animals. Plant based phytogenic feed additive being natural does not pose this serious problems. Though amla has various medicinal applications, but it is the need of hour to explore its additive values at nutritional as well as molecular level with help of various biotechnological tools and techniques. Amla has variety of phytochemical such as tannins, flavonoids, terpenoids and alkaloids are reported to indicate several pharmacological properties such as carminative, digestive, stomachic, laxative, antioxidant, antitumor, antigenotoxic and anticarcinogenic effects. It is considered to be safe herbal feed additives without any adverse effects. Further studies should be needed to standardize the inclusion levels in livestock's especially in large ruminants.

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