



## Evaluation of dietary supplementation of Aloe vera as an alternative to antibiotic growth promoters in broiler production

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### ABSTRACT

**Objective:** This study was planned to evaluate the extracts of Aloe vera (AV) at three different pH as a growth promoter, immunostimulant and coccidiostat agent in broiler production.

**Materials and Methods:** This experiment was undertaken for a period of 35 days in the Institute of Animal Sciences, Faculty of Animal Husbandry, University of Agriculture Faisalabad, Pakistan. A total of 150 day-old broiler birds (Cobb) were randomly assigned to 5 treatments with 3 replicates of 10 chicks each. Birds were offered commercial broiler starter and finisher diets supplemented with 100 mg/kg enramycin (ANT), 50 ml/liter AV of pH 3 (AV1), 50 ml/liter AV of pH 7 (AV2), 50 ml/liter AV of pH 12 (AV3) and only fresh clean water (CON).

**Results:** The results revealed that ANT and AV1 treatments caused a significantly ( $P<0.05$ ) increase in feed intake at 21 and 35 days of age and feed conversion ratio at 21 days of age. The highest weight gain was obtained by AV1 at 35 days of age ( $P<0.05$ ). Feeding diet AV1 and AV3 significantly ( $P<0.05$ ) improved the serum antibody titers against IDBV (Infectious bursal disease and Newcastle disease vaccine respectively). Fecal shedding of coccidial oocysts decreased 47.3, 39.6, 34.5 and 32.1% in AV1, AV3, ANT and AV2 respectively as compared to un-supplemented group (CON).

**Discussion:** Being an herbal product and alternative to antibiotic growth promoters AV1 treatment showed relatively better results on growth performance parameters, immune response and coccidiosis in broiler chicken.

**Keywords:** Newcastle disease; Infectious bursal disease; Immunity; Antibiotics; Growth promoters

### INTRODUCTION

Poultry production has been inundated serious problems such as poor performance, stunted growth, high feed costs, unavailability of the high quality of parent stock, diseases, and absence of well-organized marketing channel (Maqbool *et al*, 2005). The major factors influencing the poultry

production are balanced nutrition, genetic potential of breeds, and adequate health status of birds (Hussain *et al*, 2015). Feeding is about 70% of the total input cost in poultry production (Oladokun and Johnson, 2012). However, it is a matter of time to produce high quality products at the shortest possible time at the lowest cost. In the poultry industry, chemical feed additives particularly

antibiotic growth promoters (AGPs) are usually being included in chicken diet to prevent the diseases in order to improve the performance, and meat and egg production (Mile *et al.*, 2006). Excessive use of AGPs in animal diet is producing complications by developing antimicrobial resistance in pathogens (El-Hakim *et al.*, 2009). Antibiotic residues present in poultry products exhibit harmful effects on living cells and lower the quality of products. People have started to put demand on less use of AGPs in poultry especially in broiler production. As a result, the European Union (2006) has banned the use of AGP in animal feed (Castanon, 2007). In the modern era, chemical feed additives particularly antibiotics are questioned due to their undesirable effects on living cells and environment, consequently, a series of researches have started to find out the safe and effective alternatives in order to improve the performance and immune system of poultry birds. Among the conceivable options, prebiotics, probiotics, and additives from plant origin such as plant extracts, herbs and spices are considered as natural feed additives because they are safe to use, effective, and acceptable among consumers (Pourhossein *et al.*, 2015; Salim *et al.*, 2013).

Among herbal products, Aloe vera is a well-known medicinal plant. It comprises more than 75 bioactive compounds which have numerous applications and benefits (Akhtar *et al.*, 2012). They are famous for their wide range of pharmacological activities such as antibacterial, antiviral, antifungal, antiparasitic, analgesics, antioxidant and immune-stimulant properties (De Oliveira *et al.*, 2018; Yim *et al.*, 2011). AV can be used in the form of gel, powder, ethanolic extract, aqueous extract for poultry production (Babak and Nahashon, 2014).

Based on a review of the published literature, few studies were undertaken focusing on the effects of AV on growth performance parameters, and the immune response of broiler chicken (Akhtar *et al.* 2012; Darabighane *et al.* 2012; Khan *et al.*, 2014; Mmereole, 2011) but the results were inconsistent. Furthermore, standard inclusion level, best form, and comparison of AV with other medicinal herbs as a feed additive in poultry production are still unclear. In this backdrop, this study was carried out to evaluate the potential of AV as an alternative to AGPs at three different pH on performance, immune response, and coccidiosis in broilers.

## MATERIALS and METHODS

### Experimental design

The current study involved one hundred and fifty day-old broiler chicks (Cobb) purchased from the nearby hatchery which were randomly distributed to five different treatments with 3 replicates of 10 chicks each. Birds were fed either a commercial starter (ME = 2900 kcal/kg; CP = 21%) and finisher diet (ME = 3060 kcal/kg; CP = 18%) supplemented with 100 mg/kg enramycin and 60 mg/kg salinomycin (positive control), 50 ml/L AV of pH 3 (AV1), 50 ml/L AV of pH 7 (AV2), 50 ml/L AV of pH 12 (AV3) and fresh clean water (CON).

**Table 1.** Ingredients and composition of basal diet for experiment

Ingredients (%)	Starter	Finisher
Maize	50.9	58.3
Canola Meal	10.0	10.0
Sunflower Meal	20.0	20.0
Soyabean Meal	11.2	3.4
Palm oil	3.6	4.8
Limestone	1.3	1.1
Sodium Chloride	0.3	0.2
Sodium Bicarbonate	0.1	0.1
Di-Calcium Phosphate*	0.8	0.5
Lysine sulphate	0.68	0.60
Threonine	0.10	0.08
L-Isoleusine	0.03	0.03
DL-Methionine	0.24	0.15
Phytase	0.01	0.01
Vitamin Premix**	0.02	0.02
Antioxidant	0.0075	0.0075
Betain HCl	0.08	0.08
Crude protein	21	18
ME (kcal/kg)	2900	3060
Crude fiber	6	6
Calcium	0.90	0.76
Available Phosphorus	0.45	0.38
Lysine	1.32	1.05
Methionine	0.50	0.43

\*Di-Calcium Phosphate contained: 15% phosphorous and 22% calcium. \*\*Vitamin premix per kg of diet: Vitamin A, 500 IU; vitamin D3, 80.0 IU; vitamin E, 18 IU; vitamin K3, 2mg; thiamine, 150 IU; riboflavin, 300 mg; niacin, 1.5 mg; panthothenic acid, 1 mg; pyridoxine, 3mg; biotin, 0.1 mg; folic acid, 1mg; cyanocobalamin, 0.015 mg and 30mg choline chloride.

The AV extract was prepared by drying the AV leaves under shade and grinding them to make leaf powder. After that, 4-gram of leaf powder was added per liter in each separate container of HCl of

pH 3, distilled water of pH 7, NaOH of pH 12 and boiled at 80°C for 3-hours to prepare the extract. Prepared extracts of AV were filtered and cooled at room temperature. The basal diet was brought in mesh form from a commercial feed mill for starter (1-21 days) and finisher (22-35 days) periods (Table 1). Antibiotic (enramycin 100 mg/kg) was added only in ANT group. The ad-libitum provision of feed and fresh clean water was ensured to the birds throughout the experiment. A lighting program was used with 23-hours light period with 1-hour darkness. The room temperature was maintained at 95°F during the first week then decreased gradually up to 75°F and then kept constant.

#### Performance parameters

The chicken weight and feed intake (FI) in all replicates were recorded individually at 21<sup>st</sup> and 35<sup>th</sup> day of the experiment. The values of body weight gain (BWG) and FI were recorded in the starter and finisher phase and feed conversion ratio (FCR) was calculated. Mortality record in each replicate was also recorded throughout the experimental period, but it was not statistically analyzed.

#### Immunological test

In order to check the humoral response against ND and IBD vaccines, all the birds were vaccinated with ND vaccine on day 7<sup>th</sup> and 21<sup>st</sup> and also with IBD vaccine on day 12<sup>th</sup> and 21<sup>st</sup> through eye drops and drinking water. After seven days of last vaccination, blood samples were taken from three birds from each replicate through wing vein puncture. Serum antibody titers against IBD and ND vaccines were determined using haemagglutination inhibition (HI) assay previously described by (Allan and Gough, 1974) and (Hussain *et al*, 2003) respectively.

#### Parasitological procedure

Fecal samples from each treatment group were collected at day 15<sup>th</sup> and 30<sup>th</sup> for the examination of natural endoparasitic infection particularly of coccidiosis. Samples were examined using a McMaster counting chamber as previously described by (Yim *et al*, 2011). The fecal examination was performed fortnightly because the life cycle of these protozoa is one or two weeks (Akram *et al*, 2019).

#### Statistical Analysis

To analyze the results, collected data were subjected to statistical analysis using analysis of variance technique with a completely randomized design using SAS Institute (2001). Means of variables

among the different treatments were compared by Tukey's test.

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## RESULTS

The effects of all treatment groups on the growth performance parameters and immune response of broiler chickens are presented at table 2 and table 3. Treatments of ANT and AV1 significantly ( $P<0.05$ ) increased the FI at 21 and 35 days of age and FCR at 21 days of bird's age. The highest BWG was observed by AV1, which was not significantly ( $P<0.05$ ) differ from ANT at 35 days of age. FCR was tended to decrease when broilers fed with AV1 treatment. Least mortality was observed in ANT followed by AV1, AV2 and AV3 and CON during the whole experimental period. Birds receiving the AV3 and AV1 showed significantly higher ( $P<0.05$ ) antibody titer against ND and IBD vaccines respectively compared to other treatments. ANT and all AV-supplemented groups reduced the fecal shedding of coccidial oocysts as compared to CON. The groups that were supplemented with AV1, AV3, ANT and AV2 exhibited a reduction of 47.3, 39.6, 34.5 and 32.1% coccidial fecal oocyst respectively.

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## DISCUSSION

In the current study, all the treatments showed more pronounced effects at a younger age since better FCR was seen in all dietary treatments at starter phase, was not observed at finisher phase. It could be quite possible due to the fact that nutrient requirements of the older birds decreased with the increase in age and they developed an efficient digestive system (Shokri *et al*, 2017). Extracts of medicinal plants enhance the digestibility, balance the gut microflora and stimulate the secretion of endogenous digestive enzymes resulting in the increase of growth performance in poultry. Mmereole, (2011) explained that dietary supplementation of AV powder in a broiler diet possessed positive effects on BWG and FCR. It is similar to the present study in which the addition of AV in the basal diet particularly AV1 treatment resulted in improvement of FCR and BWG. Shokri *et al*, (2017), Salary *et al*, (2014), Shokraneh *et al*, (2016) also reported similar results in broiler chicken supplemented with AV and/or other herbal plants. AV contains antimicrobial substances such as anthraquinones and pyrocatechol which inhibit the activity of wide range of pathogens (Radha and Laxmipriya, 2015; Yun *et al*, 2009). These substances could play an important role in balancing intestinal

microbiota in broilers (Ahlawat and Khatkar, 2011) resulted in the improvement of productive traits.

As AV possesses antimicrobial properties, the immune response of the birds was expected to be elevated. Darabighane *et al.* (2012) reported that supplementation of AV gel at the level of 2.5% had a positive effect on antibody titer against ND vaccine in broilers. It is similar to the finding of current study that the dietary inclusion of AV can improve humoral immunity against IBD and ND vaccines. Durrani *et al.*, (2008), Elaiyaraja *et al.*, (2016), Feng *et al.*, (2011), Sharma *et al.*, (2018), Sun *et al.*, (2011) also reported that herbal products given to chicken in diet enhanced the immunities against

various diseases. It might be due to the presence of acemannan in AV containing mannose that can bind with mannose receptors present on macrophages and activate them. Additionally, It also stimulate the production of cytokines and release of nitric oxide which provide aid to birds for fighting against pathogens (Babak and Nahashon, 2014).

AV also have anti-parasitic properties both in vitro and in-vivo (Dutta *et al.*, 2008; Maphosa *et al.*, 2010). Similar to the findings of the current study, dietary supplementation of AV resulted in the reduction of fecal shedding of coccidial oocysts (Yim *et al.*, 2011).

**Table 2.** The effects of different dietary treatments on growth performance parameters of broiler in starter, finisher and overall experimental periods.

Items	Inclusion level	Initial weight	<sup>1</sup> FI		<sup>2</sup> BWG		<sup>3</sup> FCR		Mortality
		Day 1	1-21 day	22-35 day	1-21 day	22-35 day	1-21 day	22-35 day	
ANT	Enramycin*	44	1441.3 <sup>a</sup>	3591.3 <sup>a</sup>	902.3	1936 <sup>ab</sup>	1.60 <sup>a</sup>	1.86	6
AV1	50ml AV of pH 3	43.6	1429.3 <sup>a</sup>	3519.3 <sup>ab</sup>	923	1968.3 <sup>a</sup>	1.55 <sup>ab</sup>	1.82	8
AV2	50ml AV of pH 7	44	1308 <sup>b</sup>	3485.6 <sup>b</sup>	852	1762.6 <sup>b</sup>	1.54 <sup>c</sup>	1.98	9
AV3	50ml AV of pH 12	44	1335.3 <sup>b</sup>	3497 <sup>b</sup>	891	1812.6 <sup>b</sup>	1.50 <sup>bc</sup>	1.93	9
CON	Fresh clean water	43	1331 <sup>b</sup>	3480.5 <sup>b</sup>	888	1792 <sup>b</sup>	1.50 <sup>c</sup>	1.94	10
P value			0.05	0.22	0.25	0.15	0.00	0.78	
SEM		0.20	10.35	39.17	14.33	10.63	0.02	0.02	

<sup>a, b, c</sup> column means with different superscripts differ significantly within a row (p<0.05)

SEM= Standard Error of the Mean, <sup>1</sup>Feed intake, <sup>2</sup>Body weight gain, <sup>3</sup>Feed conversion ratio, \*100mg/kg

**Table 3.** The effects of different dietary treatments on immune response of broilers

Treatment Parameter	ANT	AV1	AV2	AV3	CON	SEM	P
ND	64.34 <sup>b</sup>	82.33 <sup>ab</sup>	81.31 <sup>ab</sup>	103.67 <sup>a</sup>	77.45 <sup>ab</sup>	3.77	0.000
IBD	77.67 <sup>b</sup>	99.66 <sup>b</sup>	72.56 <sup>ab</sup>	94.66 <sup>a</sup>	72.47 <sup>ab</sup>	2.68	0.021

<sup>a, b, c</sup> column means with different superscripts differ significantly within a row (p<0.05)

SEM= Standard Error of the Mean

## CONCLUSION

Dietary supplementation of Aloe vera proved satisfactory results on FCR, FI, and BWG and have the potential to be used as an alternative to AGPs in

broiler production. It also improved the antibody titer against ND and IBD vaccine. Furthermore, dietary supplementation of AV resulted in the decrease in natural infection of endoparasites especially coccidiosis in broiler. Further

investigations in different situations are expected to clarify its exact action, best form to be used and standard inclusion rate in poultry particularly broiler diets.

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