



RESEARCH ARTICLE

Effect of Administration of *Withania somnifera* on Some Hematological and Immunological Profile of Broiler Chicks

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ABSTRACT

This study was conducted to assess the performance of broiler chickens based on some hematological and immunological profile, upon inclusion of aqueous extract of *Withania somnifera* (*W. somnifera*) in their drinking water. For this purpose, 240 a-day-old broiler chicks were purchased from a local hatchery and divided into four groups i.e. WST-0, WST-I, WST-II and WST-III. Group WST-0 was kept as control, while chicks in group WST-I, WST-II and WST-III were offered in their drinking water with an extract of *W. somnifera* at 10, 20 and 30g/L for 35 days, starting from day 1 of age. Feed intake and body weight were recorded on regular basis and hematological and immunological profile of the birds was analyzed at the end of experiment. Administration of *W. somnifera* extract resulted in significantly higher feed intake and body weight of birds in all treated groups. An increase in the values of Hb, PCV and TLC was noted in the treatment groups. A non significant difference was noted in the counts of neutrophils, lymphocytes, eosinophils and monocytes in all treatment groups as compared to control. Antibodies titers against IB and IBD were significantly higher in all treatment groups as compare to the values shown by the chicks in control group. Findings of this study indicated that administration of *W. somnifera* extract to broiler chicks improve their feed intake, body weight gain, hematological profile and immunological status.

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INTRODUCTION

Withania somnifera also known as Ashwagandha or Indian ginseng, is a medicinal plant belonging to family solanaceae. This plant is cultivated and also found as natural herb in many dry and hilly areas of India and Pakistan. The main constituents of this plant are alkaloids and steroidal lactone, but the withanine, the main alkaloid found in its roots and leaves is thought to be responsible for its biological activity. The extract of this plant is a potent immune-stimulator, antioxidant and anticarcinogenic (Davis and Kuttan, 2000; Mishra *et al.*, 2000; Malik *et al.*, 2007; Sharma *et al.*, 2010).

The use of *W. somnifera* has been mainly associated to its modulatory effects on the immune system (Gautam *et al.*, 2004). It has been reported that *W. somnifera* significantly increase white blood cell and erythrocyte counts (Sham *et al.*, 2003; Manish *et al.*, 2004;

Senthilnathan *et al.*, 2006). Preparations obtained from this plant have been shown to enhance circulating antibody titer, increase the activity of lysosomal enzymes and phagocytosis (Agarwal *et al.*, 1999). Experimental animals treated with *W. somnifera* showed significant increase in hemoglobin (Hb) concentration, red blood cell (RBC) counts, white blood cell (WBC) counts, platelet counts and body weight as compared to control (Ziauddin *et al.*, 1996).

Keeping in view the importance of this plant, present study was designed to investigate its effect on hematological and immunological profile of the broiler chickens.

MATERIALS AND METHODS

Experimental design: Two hundred forty (240) one-day-old broiler chicks of approximately same weight were purchased from a local hatchery. These chicks were

divided into four equal groups i.e. WST-0, WST-I, WST-II and WST-III. Each group was further subdivided into 3 replicates, each of 20 birds. Group WST-0 was kept as control, while birds in groups WST-I, WST-II and WST-III were given an aqueous extract of *W. somnifera* at 10, 20 and 30g per liter of drinking water, respectively. All the birds were vaccinated against ND, IB and IBD at the age of day 5th, 9th, 17th, 23rd and 28th, respectively. Chicks were reared in an open sided house in pens. The broiler starter ration were fed to birds for 21 days and then changed gradually with finisher ration at the ratio of 0:4, 1:3, 2:2, 3:1 and 4:0 for remaining period. The dosing of the chicks was started from day 1 of age and lasted till the end of experiment i.e. at day 35 of age.

W. sominfera extract preparation: Fresh healthy plants of *W. sominfera* were collected from hilly areas of NWFP in summer season at flowering stage. The identification of the herb was authenticated by Department of Weed Science, NWFP Agricultural University Peshawar, Pakistan. The leaves, fruits and roots were separately air dried, and pulverized with the help of an electric grinder and were mixed at the ratio of 1:1:1. The dried powder of the mixture was subjected to extraction with water at room temperature (25°C) and boiled for two hours. The extract, so obtained was filtered through Whatman filter paper # 4 to get pure extract of the mixture. The extract WST-I, II and III were mixed in water to prepare solution for the treatment of broiler chicks.

Parameters studied: Feed intake (g) of birds in different groups was recorded on daily basis. Body weight gain (g) was determined on weekly basis. At the end of experiment i.e. day 35 of age, all the birds in each group were killed humanely and blood was collected in EDTA containing test tubes (2 mg EDTA/ 5ml blood). These blood samples were subjected to hematological analysis for Hb concentration, PCV, WBC and DLC (Benjamin, 1978). Also, blood samples were collected without addition of anticoagulant, in order to separate their sera, to be used in hemagglutination inhibition (HI) test for the estimation of antibody titer against Newcastle disease (ND) and infectious bronchitis (IB) following the method described by Alexander and Chettle (1977). Similarly, ELISA technique was used to measure antibody titers against infectious bursal disease (IBD) as described by Alkhalaf (2009).

Statistical Analysis: All the data were subjected to analysis of variance test. Means of different groups were compared using Duncan's multiple range test (Steel and Torrie, 1981). For this purpose Statistical Analysis System (SAS) was used and results were considered significantly different from one other at $P \leq 0.05$.

RESULTS

Feed intake and body weight (g) gain of the birds treated with different levels of *W. somnifera* extract have been presented in Table 1. All the groups treated with plant extract showed significantly higher values of feed intake as compared to value in group A (control). The highest value (3231.27±0.44g) was noted in groups WST-II as compared to the lowest value of 2864.91±0.89g in group A. Chicks in group WST-II showed significantly higher body weight gain as compared to the values shown by chicks in control group.

A non significant difference was noted in the Hb concentration among all groups treated with different doses of *W. somnifera* extract. The PCV value of the birds in group WST-I and WST-II were significantly higher than the values shown by their counterpart chicks in group WST-0. A significantly higher value of mean TLC was noted in the chicks from group WST-II as compared to the chicks in group WST-0, while all other groups showed non significant difference. A non significant difference was noted in the values of neutrophils, eosinophils, monocytes and lymphocytes among all treatment groups as compared to the values shown by chicks in control group.

There was non significant difference in the values of ND titer in all *W. somnifera* extract treated groups as compared to the values shown by the chicks in control group. Antibody titers against IB and IBD were significantly higher in *W. somnifera* treated groups as compared titer in control group.

DISCUSSION

In the present study, feed intake was significantly higher in chicks treated with all doses of *W. somnifera* extract. The findings of this study are in agreement with those reported by El-Deek *et al.* (2001), who reported significant increase in the feed intake of broiler birds kept on *W. somnifera* extract. This increase in the feed

Table 1: Mean values of hematological parameters, feed intake, body weight and antibody titre against ND, IB and IBD in broiler chicks feeding with different level of *Withania somnifera* extracts.

Parameters	Groups			
	WST-0	WST-I	WST-II	WST-III
Feed intake (g)	2864.91±0.89 ^b	3202.01±0.89 ^a	3231.27±0.44 ^a	3155.23±0.44 ^a
Body weight (g)	1452.13±0.89 ^b	1550.71±0.44 ^b	1736.59±0.44 ^a	1544.32±0.44 ^b
Hb (g/dl)	6.96±0.01	7.65±0.00	7.20±0.11	6.88±0.02
PCV (%)	23.00±0.09 ^c	29.16±0.00 ^a	26.50±0.04 ^{ab}	24.33±0.00 ^{bc}
TLC (×10 ⁹ cells/L)	22.17±0.00 ^b	23.25±0.00 ^b	26.58±0.00 ^a	22.33±0.00 ^b
Neutrophil (%)	40.33±0.00	35.33±0.00	30.66±0.00	37.66±0.00
Lymphocytes (%)	53.00±0.04	53.83±0.00	64.00±0.04	54.50±0.04
Eosinophil (%)	4.16±0.00	4.66±0.00	3.83±0.00	5.16±0.00
Monocytes (%)	2.50±0.08	2.50±0.08	1.50±0.08	2.66±0.00
Antibody titer against ND	6.18±0.00	6.38±0.00	6.30±0.00	6.60±0.00
Antibody titer against IB	6.01±0.04 ^b	6.58±0.01 ^a	6.58±0.01 ^a	6.75±0.00 ^a
Antibody titer against IBD	1100.0±0.89 ^b	1350.0±0.89 ^a	1350.0±0.89 ^a	1333.3±0.89 ^a

Values (Mean±SD) bearing different superscripts in a row differ significantly ($P < 0.05$).

intake, although not reported, may be due some positive effect of *W. somnifera* extract on the digestibility of feed.

W. somnifera extract resulted in the increase of body weight at all three treatment doses, i.e., 10, 20 and 30 g/L of drinking water. Sajjad (2005) and Kakar (2006) found similar effect on the weight of broiler chicks by administering 20 g of *W. somnifera*. Similarly, Wheeler (1994) and El-Deek *et al.* (2001) also noted significantly higher body weight in broiler chicks given *W. somnifera* extract as compared to untreated birds. This increase in the body weight of chick could be correlated to the increase in feed intake of the treated birds.

In an experiment, Ziauddin *et al.* (1996) found a significant increase in Hb concentration in mice given extract of *W. somnifera*. However, in the present study non significant difference was noted in the values of Hb in all three treatment levels. This difference in the response may be specie specific effects of *W. somnifera*.

High values of PCV as noted in WST-I and WST-II in the present study, may be linked with the medicinal plant extract having an effect on increasing the blood profile, which in-turn may result in increased PCV value. Feeding aqueous extract of *W. somnifera* @ 100 mg/kg/day for 15 days to laboratory animals, Gautam *et al.* (2004) noted significantly higher values of PCV. These findings and results shown by Ziauddin *et al.* (1996) are in agreement that *W. Somnifera* has boosting effect on the hematopoietic system.

The TLC values were significantly higher in the group receiving 20g plant extract water compared with control. This increase in the TLC values may be due to stimulating effect of *W. somnifera* on the bone marrow cells as has been reported by Davis and Kuttan (2000) in the mice injected (intraperitoneally) with powdered roots of *W. somnifera* at 20 mg/dose/animal for 10 days. The increase in the presence of α -esterase positive cells in the bone marrow showing the enhancement of stem cells differentiation due to *W. somnifera* is another indicator of its hematopoietic stimulatory effects.

Although, non-significantly but higher values were noted in the number of lymphocytes, eosinophils, basophiles and monocytes in the *W. somnifera* treated chicks. The increase in the number of phagocytic cells (Davis and Kuttan, 2000; Malik *et al.*, 2007) along with their increase phagocytic potential is well documented in avian, mammalian and fish species (Mishra *et al.*, 2000; Sharma *et al.*, 2010) with the treatment of *W. somnifera*.

In literature, the immune-stimulant effect of *W. somnifera* is well documented in various animal species. In the present study, increase in the titers of antibodies against IB and IBD, in all the three *W. somnifera* levels was observed. This increase in the titers of antibodies could be due to increase in the number of immunoglobulins producing cells (lymphocytes, though non significantly higher). The findings of this study are also augmented by the earlier researcher (Wheeler, 1994; Agarwal *et al.*, 1999; Sham *et al.*, 2003; Manoharan *et al.*, 2004) using the extract of this plant in different avian and mammalian models.

Conclusions

Withania somnifera extract at 20 g/L of drinking water improves the hematological parameters, especially Hb concentration, PCV and TLC in broiler chicks. Moreover, it enhances feed intake, body weight gain and immunological status of the birds. Keeping in view the beneficial effects of this plant, further studies might be launched to recommend its use in poultry to enhance its productivity.

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