



Immunomodulatory effect of *Ocimum sanctum* in broilers treated with high doses of gentamicin

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Abstract

The purpose of the study is to evaluate the immunomodulatory effect of *Ocimum sanctum* in broilers exposed to high dose of gentamicin which is common in poultry practice of TamilNadu. Two hundred and seventy day old broiler chicks of either sex were randomly divided into nine groups of 10 each with three replicates. Different doses of gentamicin (30mg/kg & 50mg/kg) single intramuscular injection, different inclusion level of *Ocimum sanctum* crude extract (1 and 2 %) in feed and their combinations were tested. The results of the study revealed that gentamicin treatment produced significant reduction in serum total protein, albumin and globulin and numerical decrease in HI titre against Newcastle vaccine. The groups treated with *Ocimum sanctum* alone showed significant increase in all the above parameters. Dose dependent increase in all the parameters were noticed in the combination groups which clearly supports the immunoprotective effect of *Ocimum sanctum*.

Keywords: *Ocimum sanctum*, gentamicin, immunomodulation, broiler

Introduction

Intensive poultry production with fast growing strains and high stocking densities are usually susceptible to infectious agents due to varied reasons and one such important reason is reduced immune potential. Antibiotics especially gentamicin is used frequently by poultry farmers at very high doses from the day of hatching to culling/marketing. The impact analysis of this practice reveals many negative effects namely failure of treatment, escalation of treatment cost and mortality in flocks some times. Many medicinal plants showing immunomodulatory activity have been used instead of drugs because of their low toxicity for the host system, adequate absorption and capability to reach the target organ without much degradation by host enzymes. *Ocimum sanctum* Linn. (Lamiaceae) commonly known as holy basil in English, Tulsi in Hindi and Tamil is an Indian medicinal plant which is known to have ethno-medical uses such as hepatoprotective, antihyperlipidaemic, myocardial salvaging and immunostimulant effect in man and animals. Hence, the present

study was carried out to explore the possible immunomodulatory effect of *Ocimum sanctum* in broilers treated with high doses of gentamicin.

Materials and Methods

Commercial day old, unsexed broiler straight run chicks (Vencob strain) belonging to a single hatch were obtained from a commercial hatchery at Namakkaland reared under standard and uniform managerial conditions throughout the experimental period of six weeks.

The birds were fed *ad libitum* with broiler starter and finisher mash free of toxins and pesticide residues. The crude extracts of the herbal plant *Ocimum sanctum* (leaves) and the commercially available gentamicin was used for the experimental study. Experimental diets containing *Ocimum sanctum* (crude extract) at 1 & 2% levels were prepared and fed to the respective treatment groups. Gentamicin was injected on 15th day at the rate of 30 and 50mg/kg body weight as single intramuscular injection. Other control birds received equal quantity of normal saline.

Two seventy commercial day old broiler chicks were weighed, wing banded and reared in battery brooders. On eighth day, the chicks were randomly divided into nine treatment groups of ten each with three replicates. The birds were subjected to respective treatments from eighth day to 42nd day at different inclusion level of the plant extract as per (Table.1).

Table 1. Experimental design		
S. No	Treatment	Experimental groups
1	T1	Normal control
2	T2	Gentamicin - 30 mg/kg
3	T3	Gentamicin - 50mg/kg
4	T4	<i>Ocimum sanctum</i> -1% level
5	T5	<i>Ocimum sanctum</i> - 2% level
6	T6	Gentamicin - 30 mg/kg + <i>Ocimum sanctum</i> - level
7	T7	Gentamicin - 50mg/kg + <i>Ocimum sanctum</i> - 1 % level
8	T8	Gentamicin - 30mg/kg + <i>Ocimum sanctum</i> - % level
9	T9	Gentamicin - 50mg/kg + <i>Ocimum sanctum</i> - % level

All the nine groups were administered with Newcastle disease vaccine (Lasota strain) at 7th and 21st day of age. Blood samples were collected at the end of fourth week and sixth week. Serum was separated and HI titre against Newcastle disease (Alexander, 1988), serum total protein, albumin and globulin (Varley, 1980) were estimated. The data were statistically analysed by completely randomized block design (Snedecor & Cochran, 1989).

Results and Discussion

The HI titre values were estimated against Newcastle disease virus in all the groups at the end of fourth week and sixth week and presented in the Table 2. At the end of fourth week and sixth week, there was no significant difference in HI titer value among all the groups. However, gentamicin control group had shown an apparently low titre value and *Ocimum sanctum* control groups recorded increase in titre value. Combinations showed dose dependent improvement in titre value which shows the immunoprotective effect of the plant. *Ocimum sanctum* modulates the humoral immune responses by acting at various levels in the immune mechanisms such

as antibody production, release of mediators of hypersensitivity reactions, and tissue responses to these mediators on the target organs (Mediratta *et al.*, 1998). Similar immunomodulating effect was reported for *Ocimum sanctum* seed oil in bovine clinical mastitis where in both the humoral and cell mediated immune responses were increased (Mukherjee *et al.*, 2005).

The serum total protein, albumin and globulin were estimated at the end of fourth and sixth week and presented in Table 3-4 respectively. Highly significant reduction in serum total protein value was observed in gentamicin control groups when compared to normal control group at the end of fourth week. However, there was no significant difference between the groups at the end of sixth week. The results of the present study are in concurrence with the observations made by Saleemi *et al.* (2009). The reduction in serum total protein might be due to inhibition of protein synthesis by gentamicin (Buss *et al.*, 1985) and moderate fluid loss through diarrhoea as observed in this study.

The group treated with combination of low level of gentamicin and high level of *Ocimum sanctum* (T8) was similar to the normal control group (T1) and *Ocimum sanctum* control groups (T4 and T5). This reversal indicates the dose dependent protective effect of *Ocimum sanctum* and could be attributed to its hepato stimulatory effect. It is a proven antioxidant, rich in phenolic compounds such as cirsilineol, circimaritin, isothymusin, apigenin and rosemeric acid and has appreciable quantity of eugenol which might have restored the serum protein level (Mode *et al.*, 2009) At four weeks of age gentamicin control groups recorded significant reduction in serum albumin level compared to all the other groups except T7 and the same was normalized at sixth week.

A highly significant decrease in serum globulin level was noticed in gentamicin control groups compared to normal control group (T1) and *Ocimum sanctum* control groups (T4 and T5) at the end of fourth week. The globulin level was high in *Ocimum sanctum*

Table 2. Effect of *Ocimum sanctum* on Haemagglutination Inhibition (log2) titre (Mean \pm S.E) in gentamicin treated broilers

Age	T1	T2	T3	T4	T5	T6	T7	T8	T9
IV week	5.16 \pm 0.30	4.33 \pm 0.42	4.83 \pm 0.48	4.33 \pm 0.42	5.33 \pm 0.33	5.00 \pm 0.26	4.50 \pm 0.43	5.33 \pm 0.21	5.00 \pm 0.26
VI week	5.17 \pm 0.31	4.33 \pm 0.42	4.83 \pm 0.48	5.17 \pm 0.40	5.33 \pm 0.33	5.00 \pm 0.26	4.50 \pm 0.43	5.33 \pm 0.21	5.00 \pm 0.26

n=6**Overall means bearing different superscripts between columns differ significantly (P \leq 0.01)

Table 3. Effect of *Ocimum sanctum* on serum total protein level (g/dl) (Mean \pm S.E) in gentamicin treated broilers

Age	T1	T2	T3	T4	T5	T6	T7	T8	T9
IV week**	5.26 ^a \pm 0.18	4.01 ^c \pm 0.10	3.91 ^c \pm 0.12	5.38 ^a \pm 0.16	5.30 ^a \pm 0.15	4.64 ^b \pm 0.14	4.49 ^{bc} \pm 0.17	5.28 ^a \pm 0.18	4.88 ^{ab} \pm 0.16
VI week	5.54 \pm 0.21	5.15 \pm 0.25	5.19 \pm 0.18	5.23 \pm 0.19	5.28 \pm 0.12	5.53 \pm 0.12	5.36 \pm 0.14	5.45 \pm 0.17	5.10 \pm 0.21

n=6**Overall means bearing different superscripts between columns differ significantly (P \leq 0.01)

Table 4. Effect of *Ocimum sanctum* on serum globulin level (g/dl) (Mean \pm S.E) in gentamicin treated broilers

Age	T1	T2	T3	T4	T5	T6	T7	T8	T9
IV week**	3.69 ^{ab} \pm 0.26	2.94 ^c \pm 0.12	2.87 ^c \pm 0.11	3.91 ^a \pm 0.19	3.76 ^a \pm 0.23	3.20 ^{abc} \pm 0.19	3.13 ^{bc} \pm 0.21	3.72 ^{ab} \pm 0.24	3.44 ^{abc} \pm 0.19
VI week	3.93 \pm 0.29	3.59 \pm 0.30	3.56 \pm 0.20	3.63 \pm 0.25	3.69 \pm 0.14	3.97 \pm 0.17	3.73 \pm 0.19	3.86 \pm 0.16	3.50 \pm 0.19

n=6**Overall means bearing different superscripts between columns differ significantly (P \leq 0.01)

control groups followed by normal control and the groups treated with high inclusion level of *Ocimum sanctum*.

High globulin level in *Ocimum sanctum* control and treatment might be due to its immunomodulatory effect which is also dose dependent (Mode *et al.*, 2009; Singh *et al.*, 2010). Further, *Ocimum sanctum* might have shown increase in total protein, albumin and globulin level by virtue of its hepatoprotective and hepatostimulant effect (Seethalakshmi *et al.*, 1982 & Prakash *et al.*, 2008).

Conclusion

The results of the present study revealed that both the doses of gentamicin produced significant reduction in serum total protein, albumin and globulin which indicates possible immunosuppressive effect. However HI titre value against Newcastle disease vaccine was normal in both the doses. *Ocimum sanctum* produced dose dependent increase in HI titre value and significant increase in serum total protein, albumin and globulin level. The combination produced dose dependent

reversal, which clearly indicates its hepatoprotective and immunomodulatory effect. Hence *Ocimum sanctum* can be added in the poultry diet during antibiotic therapy to overcome the adverse effects and enhance the immunoprotective effect.

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