



Effect of Naked Neck Gene on Immune Competence, Serum Biochemical and Carcass Traits in Chickens under a Tropical Climate

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ABSTRACT : A comprehensive study was undertaken to evaluate the effect of naked neck (Na) gene on immune competence, serum biochemical parameters and carcass quality traits in three genotypes (NaNa, Nana and nana) of the naked neck chicken under a tropical climate (Southern India). Sixty day-old chicks (20 from each genotype) were selected randomly and reared under similar environmental conditions up to eight weeks of age. The cell mediated immune (CMI) response to phytohaemoagglutinin-P (PHA-P) was significantly higher ($p \leq 0.01$) in NaNa and Nana genotypes compared to nana birds. The humoral response as measured by antibody titre to sheep red blood cells (SRBC) was also significantly higher in NaNa. The total cholesterol, LDL and VLDL cholesterol levels were significantly ($p \leq 0.01$) lower whereas HDL cholesterol level was significantly higher in NaNa and Nana compared to nana genotype. The presence of Na allele significantly increased the live weight and dressing yield, and decreased the feather cover and abdominal fat. The naked neck genotypes (NaNa/Nana) performed better than the normal (nana) siblings for almost all the traits studied. (**Key Words :** Naked Neck Gene, Immune Competence, Serum Biochemical Parameters, Carcass Traits, Tropical Climate)

INTRODUCTION

Naked neck, a phenotypic expression controlled by single dominant autosomal gene (Na) is characterized by reduced feather in neck region in chicken. The naked neck (Na) gene is incompletely dominant; the heterozygotes can be identified by a tuft of feathers on the ventral side of the neck (Scott and Crawford, 1977) whereas homozygotes have no plumage on the neck with reduced feather tract or no feather tracts (Somes, 1988). Davenport (1914) identified the naked neck gene in 20th century; Hertwig (1933) assigned the symbol 'Na' to gene. The Na gene received greater attention in the recent past in broiler production because of its association with heat tolerance (Merat, 1986; Cahaner et al., 1993; Singh et al., 2001; Lin et al., 2006) which is considered to be the most important inhibiting factor for poultry production in hot tropical climate (Horst, 1987). In broiler chickens the 'Na' gene results in a relatively higher growth rate and meat yield than

the normal birds at normal temperature and the effect is more pronounced at high temperature (Cahaner et al., 1993). Higher meat yields were reported for Na genotypes (Yunis and Cahaner, 1999; Galal and Fathi, 2001; Patra et al., 2002; Fathi et al., 2008). Naked neck gene reduces relative feather mass by 40 and 20 percent in NaNa and Nana birds respectively, thus increasing the heat dissipation. Therefore, Na birds are more tolerant to heat stress compared to their normal siblings (Merat, 1986; Patra et al., 2002). The lower feather mass increases the effective surface area for heat dissipation and increases the sensible heat loss from the neck (Yahav et al., 1998).

Introduction of major genes of tropical interest, Na, dw and F, to overcome the heat stress in hot climates and to improve the production, which will have an impact on innate immunity, thereby affecting the immune competence of chicken (Dorny et al., 2005). However, contradictory information is available with respect to influence of Na gene on immunity (Hanush et al., 2002; Kaufman, 2008). Therefore, the present study was conducted to evaluate the effect of Na gene on immune competence, serum biochemical parameters and carcass characteristics in chicken for broiler production under tropical climate.

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MATERIAL AND METHODS

Experimental population

Naked neck gene was introduced into a synthetic broiler breeder line by crossing with a homozygous Naked neck (NaNa) population for improving the broiler production under tropical environment. The base population of Naked neck line was developed after four successive generations of backcrossing and is maintained under mild selection pressure for six week body weight for the last six generations at Project Directorate on Poultry, Hyderabad, India. To avoid inbreeding, planned pedigreed mating was practiced in each generation. A total of 60 chicks, 20 chicks of both sexes (10 from each sex) from each genotype were selected randomly from a single pedigreed hatch produced from 40 sire and 120 dams. The effect of the gene was estimated by mean deviations of the NaNa and Nana from the nana genotypes using the following formula for all the traits (Fathi et al., 2008).

$$\text{Gene effect} = (\text{NaNa or Nana-nana/nana}) \times 100$$

Rearing and management

The chicks were reared upto eight weeks of age under standard management practices under deep litter system with a decreasing lighting schedule from $34 \pm 1^\circ\text{C}$ during the first week which was gradually reduced $26 \pm 1^\circ\text{C}$ by third week of age after which chicks were maintained at room temperature. The chicks were fed *ad-libitum* with broiler starter (2,900: ME, 22: CP) and finisher (3,000: ME, 20: CP) diets based on maize-soybean meal from 0-4 and 5-8 weeks of age, respectively. The chicks were vaccinated against Marek's disease, Newcastle disease, infectious bursal disease and fowl pox on 1st, 5th, 14th and 21st day, respectively. The ambient temperature ranged from 34 to 42°C during the rearing period.

Genotyping

The chicks were segregated into three genotypes based on the presence of feather on the neck region. The chicks with complete lack of feathers on the neck region was considered as homozygous dominant (NaNa), with a tuft of feathers on the ventral side of the neck as heterozygous (Nana) and with full feathers as homozygous recessive (nana) at day old age and further confirmed at 2 weeks of age.

Immune competence traits

The cellular (PHA-P) and humoral (SRBC and NDV) immune responses were studied in different set of birds during 6 weeks of age.

Cell mediated immune response

PHA-P : The birds were injected with 100 µg PHA-P solution (Bangalore Genei, Pvt. Ltd., Bangalore) in 0.1 ml sterile saline in the left wattle at 6 weeks of age. The thickness was measured with a thickness gauge (Mitutoyo) before and after 24 h of injection. The wattle swelling was calculated as the difference between the thickness of the wattle before and after the injection.

Humoral response

SRBC : Twenty birds from each genotype were injected intravenously with 0.1 ml of 0.5 percent suspension of packed sheep red blood cells (SRBC) in normal saline at 6 weeks of age. Five days later, blood was collected from wing vein of each bird. Sera were collected after two hours of incubation at room temperature and stored at -20°C . The total antibody titre was determined by haemagglutination test (Wegmann and Smithies, 1991). The reciprocal of highest dilution showing complete agglutination was expressed as titre (\log_2).

NDV : Antibody titre against NDV was determined by HI assay using 4 HA units of NDV. The highest dilution where complete inhibition of agglutination observed was read as titre (Thayer and Beard, 1998).

Serum biochemical parameters

Serum biochemical parameters like protein, total cholesterol, high density lipoprotein (HDL) cholesterol and low density lipoprotein (LDL) and very low density lipoprotein (VLDL) cholesterol was estimated in the serum samples of 20 birds from each genotype using diagnostic kits (Qualigens Pvt. Ltd, Mumbai) at 6 weeks of age. The LDL and VLDL levels were calculated by subtracting the HDL cholesterol from total cholesterol (Total cholesterol - HDL cholesterol = VLDL + LDL cholesterol).

Carcass traits

Ten male birds at 8 weeks of age were sacrificed by cervical dislocation for evaluating the carcass traits. The carcass traits such as, dressed weight, primal cuts (Legs, wings, breast and neck and back), giblets (gizzard, liver and heart) and other traits (blood, feather, head and abdominal fat) were recorded and expressed as percentage of live weight. The data were analyzed after *arc-sine* transformation of percentage values to observe the difference of means among the naked neck genotypes.

Statistical analysis

The data were subjected to a one way analysis of variance using General Linear Model (GLM) procedure in SPSS 12.0 Package. The significance of differences of means among genotypes was tested with Duncan's multiple range test (Duncan, 1955).

RESULTS

Immune competence

The cell mediated immune response as measured by PHA-P inoculation varied significantly between naked neck (NaNa/Nana) and normal (nana) genotypes. The subcutaneous dermal swelling recorded was 1.01 ± 0.1 , 0.91 ± 0.09 and 0.55 ± 0.06 mm in NaNa, Nana and nana genotypes, respectively. The antibody response to SRBC was significantly higher ($p\leq 0.01$) in NaNa/Nana compared to nana genotype (Table 1). However, no difference was observed in the ND titre among the three genotypes.

Serum biochemical parameters

The protein concentration ranged between 3.91 ± 0.10 (nana) and 4.19 ± 0.10 (Nana) with an average of 4.05 ± 0.14 g/dl and did not vary significantly among the genotypes. The total cholesterol concentration in serum was significantly ($p\leq 0.01$) lower in NaNa and Nana genotypes compared to nana genotype. The HDL cholesterol concentration was significantly higher ($p\leq 0.01$) in both naked neck genotypes compared to normal siblings whereas a reverse trend was noticed with respect to LDL and VLDL cholesterol concentration in serum (Table 2).

Carcass traits

The carcass traits in three genotypes of naked neck chicken with 'Na' gene effect are summarized in Table 3. The body weight at eight weeks of age was significantly higher ($p\leq 0.01$) in NaNa genotypes compared to nana birds. The dressing percentage of NaNa (72.39 ± 0.97) and Nana

(71.53 ± 1.37) differed significantly ($p\leq 0.01$) than nana (67.12 ± 0.66) genotypes. The percentage of blood loss was significantly higher in NaNa birds than nana genotype (Table 3). Feather proportion varied significantly among genotypes with lower proportion of feathers ($3.38\pm 0.19\%$) in NaNa birds. The head, neck and back proportion was significantly lower ($p\leq 0.01$) in NaNa and Nana birds. The proportion of leg, wing, breast muscle were not significant among the three genotypes, however, meat yield from legs (19.42 ± 0.54 g) and wings (8.46 ± 0.43 g) was higher in Nana genotypes. Breast (17.83 ± 0.26 g) meat yield was highest in NaNa genotype (Table 3). The giblets (gizzard, heart and liver) proportions did not show any significant differences among the three genotypes. The abdominal fat content was significantly lower in NaNa birds compared to other two genotypes, however, no differences on the fat proportion was found between Nana and nana genotypes (Table 3).

The presence of Na gene increased the body weight and dressing percentage by 5.4 and 7.9 and 1.6 and 6.6% in homozygous and heterozygous conditions, respectively. In primal cuts, the Na gene had positive effect on breast muscle yield and negative effects in all other traits (legs, wings and neck and back) in NaNa, whereas, positive effect for all except neck and back in Nana genotype. Compared to normal birds, the Na allele reduced the plumage cover by 36.9 and 15.7% in NaNa and Nana condition, respectively. The Na gene reduced the abdominal fat considerably by 52.3% and 21.6% in homozygous and heterozygous conditions, respectively. Head proportion also had similar findings but blood proportion had positive effect of the gene (Table 3).

Table 1. Immunocompetance parameters in three naked neck genotypes

Parameter	Genotype			Prob	Overall	Gene effect	
	NaNa	Nana	nana			NaNa	Nana
N	20	20	20		60		
PHAP (mm)	1.01 ± 0.10^a	0.91 ± 0.09^a	0.55 ± 0.06^b	0.001	0.81 ± 0.05	83.64	65.45
SRBC (\log_2)	7.00 ± 0.29^a	6.88 ± 0.65^a	4.62 ± 0.38^b	0.000	6.07 ± 0.35	51.52	48.92
ND titre (\log_2)	3.81 ± 0.40	3.77 ± 0.29	$3.15\pm 0.0.28$	0.251	3.55 ± 0.19	20.95	19.68

Means with different superscripts within a row differ significantly ($p\leq 0.01$).

Table 2. Serum biochemical parameters in naked neck chicken genotypes

Parameter	Genotype			Prob	Overall	Gene effect	
	NaNa	Nana	nana			NaNa	Nana
N	20	20	20		60		
Protein (g/dl)	3.98 ± 0.11	4.19 ± 0.13	3.91 ± 0.10	0.330	4.05 ± 0.14	1.79	7.16
Total cholesterol (mg/dl)	141.80 ± 2.51^b	151.38 ± 3.92^b	167.35 ± 5.26^a	0.001	153.51 ± 3.05	-15.27	-9.54
HDL cholesterol (mg/dl)	112.67 ± 3.90^a	109.78 ± 3.36^a	94.25 ± 2.92^b	0.002	105.57 ± 2.47	19.54	16.48
LDL and VLDL Cholesterol (mg/dl)	38.72 ± 5.37^b	47.54 ± 3.86^{ab}	57.56 ± 6.58^a	0.046	47.94 ± 3.34	-32.73	-17.41

Means with different superscripts within a row differ significantly ($p\leq 0.01$).

Table 3. Carcass quality traits in three genotypes of naked neck chicken expressed in percentage of live weight

Parameter	Genotype				Overall	Gene effect	
	NaNa	Nana	nana	p		NaNa	Nana
N	10	10	10		30		
Live weight (g)	1,943.1±44.06 ^a	1,873.83±62.78 ^{a,b}	1,844.1±69.24 ^b	0.012	1,886.19±32.6	5.37	1.61
Dressing percentage	72.39±0.97 ^a	71.53±1.37 ^a	67.12±0.66 ^b	0.005	70.42±0.73	7.85	6.57
Primal cuts							
Legs	18.58±0.50	19.42±0.54	18.91±0.68	0.601	19.00±0.33	-1.75	2.70
Wing	7.39±0.65	8.46±0.43	8.11±0.22	0.191	8.01±0.27	-8.88	4.32
Breast	17.83±0.26	16.84±0.54	16.76±0.65	0.275	17.12±0.30	6.38	0.48
Neck and back	15.36±1.04 ^b	17.78±0.53 ^a	18.78±0.87 ^a	0.019	17.33±0.52	-18.21	-5.32
Giblets							
Gizzard	2.16±0.12	2.25±0.06	2.34±0.14	0.512	2.25±0.06	-7.69	-3.85
Liver	2.68±0.15	2.65±0.14	2.50±0.09	0.557	2.60±0.07	7.20	6.00
Heart	0.49±0.03	0.46±0.02	0.44±0.02	0.264	0.46±0.01	11.36	4.55
Other traits							
Blood	7.38±0.23 ^a	6.21±0.33 ^b	5.19±0.32 ^c	0.000	6.26±0.23	42.20	19.65
Feather	3.38±0.19 ^b	4.52±0.39 ^a	5.36±0.34 ^a	0.001	4.42±0.23	-36.94	-15.67
Head	2.44±0.13 ^b	2.67±0.15 ^b	3.05±0.04 ^a	0.007	2.72±0.08	-20.00	-12.46
Abdominal fat	0.55±0.12 ^b	0.91±0.01 ^a	1.16±0.10 ^a	0.003	0.87±0.08	-52.59	-21.55

Means with different superscripts within a row differ significantly ($p \leq 0.01$).

DISCUSSION

Immune competence traits

The findings of the present study suggested that Na allele in homozygous and heterozygous condition increased the CMI response to PHA-P and antibody titres to SRBC compared to their normal sibs. El Safty et al. (2005) observed significant increase of wattle swelling in naked neck hens a measure of CMI compared to their normal feathered hens similar to the present findings. Higher CMI response in NaNa and Nana genotypes compared to normal siblings are also reported in literature (Patra et al., 2004; Fathi et al., 2005; Fathi et al., 2008; Galal, 2008). Though SRBC and NDV responses are humoral in nature, the higher antibody titres to SRBC might be because of CMI dependent response, therefore the variation among the genotypes. The findings of the present study further suggested that immune competence traits can be used in selection programme in naked neck birds for improving the general disease resistance as naked neck genotypes have better immunity than the normal genotypes.

Serum biochemical parameters

The Na gene reduced the total cholesterol concentration by 15.3 and 9.5% and LDL cholesterol concentration by 32.7 and 13.4% in NaNa and Nana genotypes respectively. The Na allele increased the HDL cholesterol level by 19.5 and 16.5% in homozygotes and heterozygotes, respectively.

The total cholesterol concentration was significantly ($p \leq 0.01$) lower in both genotypes of naked neck compared to their normal sibs. The serum cholesterol levels are directly proportional to muscle cholesterol levels, therefore, it can be stated that the naked neck genotypes studied, have lower muscle cholesterol concentration. The HDL concentration was significantly higher in naked neck genotypes, which is desirable and designated as good cholesterol for its ability to mobilize cholesterol for body metabolism thus reducing the cholesterol concentration in the body. The triglycerides, VLDL and LDL are the indicators of fat deposition (Griffin et al., 1982). The VLDL and LDL concentration were significantly lower in NaNa and Nana birds indicating lower fat deposition in naked neck broilers. Similar findings were observed by Patra et al. (2004) and Zein-El-Dein et al. (1984).

Carcass traits

The significant higher body weights of NaNa and Nana birds observed in the present study indicated the effects of Na gene on body weight either in homozygous or heterozygous state. Higher body weights for naked neck genotypes were also reported in literature (Cahanar et al., 1994; Galal and Fathi, 2001; Fathi et al., 2008). The dressing percentage was significantly higher in two naked neck genotypes than their normal siblings, which can be attributed to higher body weights and less losses due to feather in naked neck birds. The higher meat yield due to

the presence of Na gene is well established (El Attar and Fathi, 1995; Deeb and Cahaner, 1999; Yalcin et al., 1999; Fathi and Galal, 2001; Fathi et al., 2008).

The increased proportion of muscle in pectoral region in Na genotypes might be due to the availability of higher levels dietary protein for muscle development with less protein requirement for plumage feather development (Merat, 1990). El Attar and Fathi (1995) observed significantly higher breast yields in naked neck genotypes under high ambient temperatures. Similarly, better meat yield in naked neck genotypes was also reported by Yalcin et al. (1997). The higher meat yield was attributable to high protein availability in naked neck birds which lead to high muscle fibre formation (Merat, 1990).

The giblets proportion was not significantly varied among genotypes. However, liver and heart weights were more in naked neck genotypes, while gizzard was heavier in normal birds. The heavier liver and heart in naked neck genotypes may be correlated with high metabolic rate and high blood pumping from the heart to meet the high metabolic needs of the birds. The significant higher blood proportion in NaNa and Nana birds also confirms the association between heart and liver in naked neck chicken.

The abdominal fat percentage in relation to live weight was significantly lower in both NaNa and Nana genotypes compared to the normal birds. The significant effect Na gene in reducing the subcutaneous and intra muscular fat deposition is well documented (Cahaner et al., 1993; Raju et al., 2004; Fathi et al., 2008). The body fat content is negatively correlated with heat tolerance (Macleod and Hocking, 1993) which substantiates the present findings of less fat in naked neck birds. Patra et al. (2002) reported higher fat percentages in naked neck genotypes which were not significant among the genotypes. The low level of average abdominal fat percentage (0.87 ± 0.08), observed in the present study in naked neck is highly desirable for improving the carcass quality and minimizing the deposition of energy. The decrease in abdominal fat in naked neck chicken may be due to the varied insulation effects of less plumage cover and utilization of a higher proportion of energy on thermoregulation leading to less body fat deposition (Merat, 1986).

CONCLUSIONS

The performance of naked neck genotypes (NaNa and Nana) was superior in all the traits, i.e, immune competence, serum biochemical parameters and carcass quality traits, evaluated in the present study because of favourable expression of Na gene under hot climatic conditions due to better adaptability of the birds. The naked neck gene can be a viable alternative for broiler production especially in the

tropical countries where summer temperatures are very high.

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