Genetic Studies on Immuno-competence Traits in White Leghorn Chicken

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Abstract
The present investigation was carried out to assess the immunocompetence traits and their inter-relationship in four White Leghorn strains. Significant differences among genetic groups were observed for antibody titre against SRBC and NDV. Mean anti-SRBC titre, anti NDV titre and response to PHA-P (mm) ranged from 4.729 ± 0.142 to 5.803 ± 0.118, 10.033 ± 0.056 to 11.045 ± 0.049 and 0.133 ± 0.075 to 0.527 ± 0.164, respectively. Heritability estimates of antibody titre against SRBC, NDV and response to PHA-P were low to medium. The heritability from sire x dam component of variance varied from 0.188 ± 0.087 to 0.268 ± 0.173 for anti-SRBC titre, from as low as 0.108 ± 0.104 in WI as high as 0.434 ± 0.226 for anti-NDV titre and from 0.133 ± 0.075 to 0.217 ± 0.143 for response to PHA-P. The genetic, phenotypic and environmental correlations among the immunocompetence traits ranged from 0.122 ± 0.14 to 0.403 ± 0.286, -0.010 to 0.09 and -0.188 to 0.683, respectively across all White leghorn strains. The positive association among the immune parameters suggested the possibility of simultaneous improvement in both humoral and cell mediated immunity.

Key words: Immuno-competence, White Leghorn, SRBC, NDV, PHA-P, Heritabilities, Correlations.

1. Introduction
The poultry industry has been incurring substantial losses through mortality and morbidity due to infectious diseases worldwide. Conventional control of poultry diseases involves high-cost sanitary measures, vaccination programs, and medical treatments. Moreover, there is increasing public objection to drug residues in poultry products. Thus, breeding chickens for higher immunocompetence and disease resistance provides an alternative approach to securing poultry viability. The humoral, cell mediated immunity and phagocytic index may be put together along with the growth and/or egg production traits in an index for selection of parents. The progress may be slow, but this will produce the lines that can survive under adverse climatic conditions (Yang and Jiang, 2005). Selection for immunocompetence was suggested as an indirect method of improving disease resistance (Gavora and Spencer, 1983; Van der Zijpp, 1983). Results of the selection experiments in the past revealed that selection for increased antibody production to one antigen such as SRBC improved the responses to several antigens (Siegel and Gross, 1980). However, effects of selection for traits such as egg production on immune traits or the association of immune traits with production traits need to be studied. The reports on genetic correlation of immunocompetence traits with growth and production traits in layers are very scanty in literature. Therefore, the goal of the current study was to evaluate the immunocompetence measurements and their inter-relationship in four White Leghorn strains.

2. Materials and Methods

2.1 Chicken Populations
The three strains of White Leghorn (IWH, IWI, and IWK) and the control population maintained at Directorate of Poultry Research, Hyderabad utilized in the present study were procured from different sources and had different histories of selection. There was no...
selection for any of the immune-competence traits in these chicken populations.

General immunity traits such as antibody response against SRBC as total immunoglobulin at 5 days post injection of SRBC (at 24 weeks of age), humoral immunity to Newcastle disease virus vaccine (at 28 weeks of age) and response to PHA-P (at 30 weeks of age) for assaying cell mediated immune response were evaluated in pure lines of White Leghorn by using the following procedures.

2.2 Antibody Response to Sheep Red Blood Cells (SRBC)

A total of 1214 female birds from IWH (337), IWI (301) and IWK (335) strains and control population (241) were utilized to study the antibody response to SRBC. Sheep RBC is a complex, non pathogenic T-cell dependent antigen which initiates general immune responsiveness (Gavora, 1990). The method described by Siegel and Gross (1980) with slight modifications was followed for assessing the immune response to SRBC. About 5 ml of blood from healthy sheep was collected into vacutainer tubes containing EDTA, the RBCs were washed in PBS and a 0.5% suspension of SRBCs was prepared. Each bird was injected with 0.1 ml of SRBC suspension intravenously. At 5 days post injection, about 1 ml blood was collected from wing vein from which serum was obtained. Titre of antibodies generated against SRBC was determined by haemagglutination (HA) test. The reciprocal of highest dilution showing complete agglutination was expressed as titre (n). The titre was transformed into log₂ values for further analysis.

2.3 Antibody Response to Newcastle Disease Virus (NDV)

A total of 1046 birds consisting of IWH (289), IWI (251), IWK (310) and control population (196) were included in the experiment. The birds were vaccinated at 10 weeks of age using Newcastle Disease killed vaccine. The blood samples were collected from birds at 25-30 weeks of age. The haemagglutination inhibition titre using 4 HA units of NDV was taken as indicative of humoral immunity against NDV vaccine and titres were estimated.

2.4 Cell Mediated Immune Response

A total of 448 female birds from IWH (90), IWI (115), IWK strains (137) and control population (106) were utilized to estimate the In vivo T cell mediated immune response to mitogen (PHA-P) as per Cheng and Lamont (1988) by using the wattle response to mitogen at the age of 30 weeks. The birds were injected with 0.1 mg of PHA-P dissolved in 0.1 ml phosphate buffer saline intra-dermally into the left wattle of the birds while 0.1 ml of PBS was injected in to the right wattle as control. The thickness of wattle was measured using a thickness gauge (Mitutoyo) and the wattle swelling (cell mediated immune response) in millimeters was calculated as the difference in the increase in thickness of left wattle (injected with PHA-P) and the right wattle (injected with PBS) as given below.

Response = (Post Injection – Pre Injection) – (Post PBS – Pre PBS)

Where,
Post Injection = Thickness of wattle 24 hours after injection of PHA-P
Pre Injection = Thickness of left wattle before injection
Post PBS = Thickness of right wattle 24 hours after injection of PBS
Pre PBS = Thickness of right wattle before injection.

2.5 Statistical Analysis

Since the data were collected from birds produced in six hatches, it was adjusted for significant hatch effects as per Harvey (1990) and the hatch corrected data were utilized for further statistical analysis. Heritabilities and genetic, phenotypic and environmental correlations were estimated by full sib correlation method using Mixed model least squares and maximum likelihood (LSMLMW) computer program (Harvey, 1990). The statistical model for the estimation of heritability of various traits studied was as follows.

\[ Y_{ik} = \mu + s_i + d_j + e_{ijk} \]

Where,
\[ Y_{ik} = \text{measurement of a trait on } k^{th} \text{ progeny belonging to } j^{th} \text{ dam mated to } i^{th} \text{ sire.} \]
\[ \mu = \text{Overall mean} \]
\[ s_i = \text{effect of } i^{th} \text{ sire} \]
\[ d_j = \text{the effect of } j^{th} \text{ dam mated to the } i^{th} \text{ sire} \]
\[ e_{ijk} = \text{uncontrolled environmental and genetic deviations attributable to the individuals} \]

The heritability was computed from sire, dam and sire + dam components of variance as given below.

\[ h^2_S = 4 \times (\sigma^2_S / (\sigma^2_S + \sigma^2_D + \sigma^2_W)) \]
\[ h^2_D = 4 \times (\sigma^2_D / (\sigma^2_S + \sigma^2_D + \sigma^2_W)) \]
\[ h^2_{S+D} = 2 \times (\sigma^2_S + \sigma^2_D / (\sigma^2_S + \sigma^2_D + \sigma^2_W)) \]

Estimation of co-efficient of correlation
Table 1: Analysis of variance of immunocompetence traits in White Leghorns

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>d.f</th>
<th>SRBC</th>
<th>NDV</th>
<th>PHAP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between strains</td>
<td>3</td>
<td>56.474(^{**})</td>
<td>46.785(^{**})</td>
<td>0.096</td>
</tr>
<tr>
<td>Between hatches</td>
<td>3</td>
<td>3.790</td>
<td>0.622</td>
<td>0.070</td>
</tr>
<tr>
<td>Error</td>
<td>3.759 (1207)</td>
<td>0.558 (1039)</td>
<td>0.070 (441)</td>
<td></td>
</tr>
</tbody>
</table>

*Figures in the parentheses indicate error degrees of freedom; \(^{**}\) Significant (P<0.01)*

Table 2: Least squares means for immunocompetence traits in White Leghorns

<table>
<thead>
<tr>
<th>Trait</th>
<th>IWH Mean</th>
<th>SE</th>
<th>IWI Mean</th>
<th>SE</th>
<th>IWK Mean</th>
<th>SE</th>
<th>Control Mean</th>
<th>SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>SRBC titer</td>
<td>5.803(^{a})</td>
<td>0.118</td>
<td>5.465(^{b})</td>
<td>0.133</td>
<td>4.776(^{c})</td>
<td>0.110</td>
<td>4.729(^{c})</td>
<td>0.142</td>
</tr>
<tr>
<td>NDV titer</td>
<td>11.045(^{a})</td>
<td>0.049</td>
<td>10.033(^{d})</td>
<td>0.056</td>
<td>10.721(^{b})</td>
<td>0.044</td>
<td>10.393(^{c})</td>
<td>0.61</td>
</tr>
<tr>
<td>Response to PHA-P (mm)</td>
<td>0.719</td>
<td>0.030</td>
<td>0.720</td>
<td>0.030</td>
<td>0.743</td>
<td>0.024</td>
<td>0.796</td>
<td>0.030</td>
</tr>
</tbody>
</table>

*Means with different superscripts in each row differ significantly*

The genetic, phenotypic and environmental correlations between various traits were also estimated from S+D components of variance using Least squares and maximum likelihood computer program (Harvey, 1990) based on following formulae.

a. Genetic Correlations:

\[
r_G = \frac{\text{Cov}_S + \text{Cov}_D}{\sqrt{\sigma^2_S(x) + \sigma^2_D(x)} \sqrt{\sigma^2_S(y) + \sigma^2_D(y)}}
\]

b. Phenotypic Correlations:

\[
r_P = \frac{\text{Cov}_W + \text{Cov}_S + \text{Cov}_D}{\sqrt{\sigma^2_W(x) + \sigma^2_S(x) + \sigma^2_D(x)} \sqrt{\sigma^2_W(y) + \sigma^2_S(y) + \sigma^2_D(y)}}
\]

c. Environmental Correlations:

\[
r_E = \frac{\text{Cov}_W + \text{Cov}_S + \text{Cov}_D}{\sqrt{\sigma^2_W(x) - \sigma^2_S(x) - \sigma^2_D(x)} \sqrt{\sigma^2_W(Y) - \sigma^2_S(Y) - \sigma^2_D(Y)}}
\]

3. Results and Discussion

3.1 Means

The details of immunocompetence traits in White Leghorns were presented in (Table 1-2). Significant differences in anti SRBC titre and anti NDV titre were observed among the genetic groups studied, which agreed well with the reported literature (Miller et al., 1992; Pinard et al., 1992; Saxena 1993; Kundu et al., 1999; Jayalaxmi, 2008). In the present study no significant strain differences were observed between the genetic groups in CMI response to PHA-P injection which is in agreement with the reports of (Santhosh, 1999; Jayalaxmi, 2008), while Kean et al. (1994) reported significant strain/line differences.

The mean anti SRBC titre ranged from 4.729 ± 0.142 in control population to 5.803 ± 0.118 in IWH strain. These results are similar to the means reported by Jayalaxmi (2008), which ranged from 4.744 ± 0.093 in control population to 5.493 ± 0.118 in IWH. However, the anti SRBC titres in the present study were lower than other reports on different chicken populations (Vasu et al., 2004; Sanjeevkumar and Kataria, 2005; Chatterjee et al., 2007) and higher than the observations of Singh et al. (2009). The titres ranged from 10.033 ± 0.056 in IWI strain to 11.045 ± 0.049 in IWH and were similar to those reported by (Shivakumar and Kumar, 2005; Jayalaxmi, 2008). Lwelamira and Katule (2004) reported significant differences between genetic groups in the antibody response against NDV. The mean increase in thickness ranged from 0.719 ± 0.030 to 0.796 ± 0.030 mm, which is comparable to that reported by (Reddy et al., 2005; Chatterjee et al., 2007; Singh et al., 2009), while Jayalaxmi (2008) reported lower means. Kean et al. (1994) reported a higher response than in the present study ranging from an increase in thickness of 1.10 to 1.658 mm. The higher response in the report reviewed may be because of the selection for immunocompetence for 7 generations.

3.2 Heritability Estimates

The details of heritability estimates for immunocompetence in White Leghorns were presented in (Table 3). The heritability of anti SRBC titre estimated from dam component was higher than that estimated from sire component in IWK strain, while the \(h^2_e\) was higher than \(h^2_d\) in other populations. Vasu et al.
Table 3: Heritability estimates for immunocompetence traits in White Leghorns

<table>
<thead>
<tr>
<th>Trait</th>
<th>IWH</th>
<th>IWI</th>
<th>IWK</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$h^2_S$</td>
<td>$h^2_D$</td>
<td>$h^2_{S+D}$</td>
<td></td>
</tr>
<tr>
<td>SRBC titer</td>
<td>0.524 ± 0.137</td>
<td>0.218 ± 0.055</td>
<td>0.187 ± 0.066</td>
<td>0.414 ± 0.251</td>
</tr>
<tr>
<td>NDV titer</td>
<td>0.242 ± 0.118</td>
<td>0.189 ± 0.081</td>
<td>0.180 ± 0.113</td>
<td></td>
</tr>
<tr>
<td>Response to PHA-P</td>
<td>0.148 ± 0.104</td>
<td>0.155 ± 0.092</td>
<td>0.204 ± 0.086</td>
<td></td>
</tr>
</tbody>
</table>

# - Not estimable

Table 4: Genetic (Above diagonal), Phenotypic (below diagonal) and environmental (below diagonal in brackets) correlation coefficients for immunocompetence traits in White Leghorns

<table>
<thead>
<tr>
<th>Particulars</th>
<th>SRBC titer</th>
<th>NDV titer</th>
<th>Response to PHA-P</th>
</tr>
</thead>
<tbody>
<tr>
<td>IWH</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SRBC titer</td>
<td>-0.122 ± 0.014</td>
<td>0.223 ± 0.123</td>
<td></td>
</tr>
<tr>
<td>NDV titer</td>
<td>-0.066 (0.519)</td>
<td>0.338 ± 0.184</td>
<td></td>
</tr>
<tr>
<td>Response to PHA-P</td>
<td>0.054(-0.188)</td>
<td>-0.058 (-0.301)</td>
<td></td>
</tr>
<tr>
<td>IWI</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SRBC titer</td>
<td>0.097 ± 0.516</td>
<td>-0.224 ± 0.175</td>
<td></td>
</tr>
<tr>
<td>NDV titer</td>
<td>-0.010 (0.683)</td>
<td>0.403 ± 0.286</td>
<td></td>
</tr>
<tr>
<td>Response to PHA-P</td>
<td>-0.062 (0.097)</td>
<td>0.072 (-0.429)</td>
<td></td>
</tr>
<tr>
<td>IWK</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SRBC titer</td>
<td>0.268 ± 0.117</td>
<td>0.236 ± 0.155</td>
<td></td>
</tr>
<tr>
<td>NDV titer</td>
<td>0.122 (0.541)</td>
<td>#</td>
<td></td>
</tr>
<tr>
<td>Response to PHA-P</td>
<td>0.071(-0.356)</td>
<td>0.091 (-0.208)</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SRBC titer</td>
<td>-0.323 ± 0.146</td>
<td>#</td>
<td></td>
</tr>
<tr>
<td>NDV titer</td>
<td>-0.118 (0.643)</td>
<td>#</td>
<td></td>
</tr>
<tr>
<td>Response to PHA-P</td>
<td>0.045 (0.322)</td>
<td>-0.053 (0.260)</td>
<td></td>
</tr>
</tbody>
</table>

# - Not estimable

(2004) and Jayalaxmi (2008) also reported similar estimates in White Leghorns. The heritability estimated from sire + dam component of variance varied from 0.188 ± 0.087 to 0.268 ± 0.173 indicating that it is low to moderately heritable. The present findings are in agreement with the reports of (Bovenhuis et al., 2002; Nath et al., 2005), while Jayalaxmi (2008) reported moderate to high estimates. The results of the present study and reported literature indicated that anti SRBC titre is controlled by both additive and non-additive types of gene action. Siegel and Gross (1980) suggested both additive and non-additive genetic control for immune response to SRBC, which is also reflected in the present study. Hence, selection may be useful for improvement of humoral immunity.

The heritability estimated from sire + dam component of variance ranged from as low as 0.180 ± 0.113 to 0.242 ± 0.118. Jayalaxmi (2008) reported very low (0.003 ± 0.676 in IWK strain) as well as very high (0.982 ± 0.394 in control population) estimates. Lwelamira and Katule (2004) reported heritabilities ranging from 0.27 ± 0.06 to 0.39 ± 0.09 in different genetic groups. Lwelamira et al. (2008) reported heritability values of 0.27 ± 0.06 and 0.29 ± 0.05 in Kuchi and Tanzania medium chicken lines of Tanzania. The high heritability obtained in IWH and IWK strains in the present study indicated that good immune response could be achieved through selection in these two strains.

The heritability estimates of antibody titre against PHA-P ranged from 0.226 ± 0.097 to 0.527 ± 0.164, 0.148 ± 0.104 to 0.204 ± 0.086 and 0.133 ± 0.075 to 0.217 ± 0.143 when estimated from sire, dam and sire ± dam components of variances respectively.
Jayalaxmi (2008) reported higher heritability estimates, while Kean et al. (1994) reported lower estimates for response to PHA-P.

3.3 Correlations

The genetic correlation (Table 4) of anti SRBC titre with anti NDV titre was found to be negative in IWH and control population and positive in IWI and IWK with a range -0.122 ± 0.014 to 0.268 ± 0.117. The genetic correlation of anti SRBC titre with CMI response to PHA-P was positive in IWH (0.223 ± 0.123) and IWK (0.236 ± 0.155) and negative in IWI (-0.224 ± 0.175) strain. The genetic correlation of anti NDV titre with CMI response to PHA-P was positive in IWH (0.338 ± 0.184) and IWI (0.403 ± 0.286) strains. Similar findings were reported by Jayalaxmi (2008).

The phenotypic correlations (Table 4) of anti SRBC titre with anti NDV titre were negative in three strains except in IWK and positive with CMI response to PHA-P in three strains except in IWI. However, the phenotypic correlations of anti NDV titre with CMI response to PHA-P were positive in IWI and IWK and negative in IWH and control population. Contrarily, Shivakumar and Kumar (2005) and Jayalaxmi (2008) reported positive phenotypic correlations.

4. Conclusions

Significant genetic group differences were observed for immunocompetence traits. Moderate to high heritability estimates of immunocompetence traits suggested that there is a good scope for improvement of generalized immunity. The positive association among the immune parameters studied suggested the possibility of simultaneous improvement in both humoral and cell mediated immunity.

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References


