Studies on Pathology of Chronic Inflammation Induced by *Escherichia coli* in Chicken Skin

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**Abstract**

The study was undertaken to investigate the nature of chronic inflammatory response induced by *Escherichia coli* in white Leghorn chickens. Inflammation was induced by intradermal injections of known concentration of *E. coli* at various time intervals and the pattern of emigration of leukocytes was recorded. In the early stage, emigration of heterophils, basophils and monocytoid cells were observed. Degranulating mast cells and macrophages were noticed on day 3 and their number increased subsequently. The inflammatory reactions were followed by reparative changes which were evidenced by the presence of mature fibroblasts and marked collagen content from day 12 onwards.

**Key words**: Angiogenesis, chronic inflammation, fibroplasia, leukocytes.

Migration of leukocytes to the site of injury is a complex mechanism. Leukocytes migrate from vascular site to the damaged tissue to destroy the potential causative agent. The cellular pattern of migration is typically different in acute and chronic inflammation. They have been studied extensively in mammals (Luster *et al.*, 2005) but limited work has been done in avian species. The present work was designed to study the cellular response in chronic inflammation in chicken.

**Materials and Methods**

For the experiment, twelve, white Leghorn chicken of both sexes, aged 10-16 weeks were equally divided into experimental and control groups. For induction of inflammation, Freeze dried culture of *E. coli* was procured and pure colonies were obtained. The bacterial suspension was then prepared from these isolates. One day before injecting the *E. coli* suspension, feathers from lateral thoracic region were plucked and skin cleaned aseptically. One lesion each of any three different time intervals were produced to induce inflammation by giving intradermal inoculation of 0.02 ml of *E. coli* suspension. Same procedure was followed for the birds of control group using non pyrogenic normal saline. Six lesions of each time interval, as shown in Table I, were obtained. Area of skin bearing the lesion was excised and processed for sequential histopathological study (Katiyar *et al.*, 1993). Tissue leukocytosis was assessed by quantifying the number of leukocytes using high power (X 400) in five representative fields.

**Results and Discussion**

Most studies on avian inflammatory process have been restricted to acute inflammation where as chronic inflammatory response remains relatively unexplored. The present work was therefore focused to study the leukocytic emigration and histopathological changes during chronic inflammation induced by *E. coli*. It was found that in early stages (day 3 to 9) emigration of heterophils, basophils and monocytoid cells were predominant. Heterophils were not observed after 3rd day, whereas, monocytoid cells-and basopils were present in the lesion until 9th day (Table I). Earlier, investigators (Vegad, 2007; Jain *et al.*, 1996) have reported the presence of heterophils in acute inflammatory response from 30 minutes to 3 days. Because of their short half life, the emigrated heterophils do not survive for long.

The monocytoid were observed from the early stages. These cells persisted until the maturation of granulation tissue, and were the predominant cells. Other workers have recorded...
marked monocytic infiltration during the later stages of acute inflammation (Jain et al., loc. cit; Gabay, loc cit). In mammals also monocytes participate in the later stages of inflammation partly because factors causing their recruitment play a role in the later stages, and partly also because their life span ranges from weeks to months.

An insignificant infiltration of basophils was noted in the early stages of chronic inflammatory response (Table I). Degranulating mast cells were also noticed at 3 day stage. The presence of basophils in early stages of acute inflammation has been reported by earlier workers using various stimuli as reviewed by Vegad and Katiyar (1995). Their report suggested that basophils may be playing a specific role in the chemical mediation of acute inflammation.

Sequential assessment of monocytoid cell infiltration revealed highest number of cells in the early stages of the present investigation. Subsequently their number decreased gradually up to 18 days. The majority of monocytoid cells emerging from blood vessels showed a considerable change in their morphology. After emigration, they undergo many structural alterations eventually becoming macrophages and their size become larger with time. As described in mammals, the macrophage activation is a complex process (Vegad, loc. cit; Yadav et al., loc cit).

In the present study, formation of perivascular lymphoid aggregates was conspicuous which were observed from 3 days onwards. Apart from the compact lymphoid aggregates, diffuse lymphocytic infiltration was also noticed. This could be due to mucopolysaccharides liberated by the degranulation of basophils which might have inhibited the spread of lymphocytes and made them compact perivascularly.

The distinct multinucleated giant cells were found from 3 days onwards. Sinha et al. (1987) has also described giant cell formation during the late stage of acute avian inflammation using different stimuli. This is marked in contrast to the situation occurring in mammals that suggest the avian species in general may be more prone to giant cell formation than mammals (Kumar et al., 1992).

The inflammatory reaction is followed by process of healing as evidenced by the process of fibroplasia, angiogenesis and collagen content from 12 day onwards. These changes were noticed from 3 days onwards. Since from the beginning, there was a heavy influx of monocytoid cells and their transformation into macrophages in the later stages, growth factors such as FGF (Fibroblasts Growth Factor) and fibrogenic cytokines such as IL-I (Interleukin-I) and TNF (Tumour Necrosis Factor) may have been produced at the injured site (Kumar et al., loc cit). Therefore, it is suggested that in the chicken monocytoid cells not only play a functional role in the process of inflammation but also in the healing and repair of damaged tissue.

<table>
<thead>
<tr>
<th>Time interval</th>
<th>Heterophils</th>
<th>Monocytoid cells</th>
<th>Basophils*</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 days</td>
<td>5.26±0.60</td>
<td>18.40±2.54</td>
<td>0.93±0.15</td>
<td>24.59±3.29</td>
</tr>
<tr>
<td>6 days</td>
<td>00</td>
<td>18.30±2.68</td>
<td>0.60±0.15</td>
<td>18.90±2.83</td>
</tr>
<tr>
<td>9 days</td>
<td>00</td>
<td>21.43±3.31</td>
<td>0.30±0.08</td>
<td>21.73±3.39</td>
</tr>
<tr>
<td>12 days</td>
<td>00</td>
<td>13.63±2.00</td>
<td>00</td>
<td>13.63±2.00</td>
</tr>
<tr>
<td>15 days</td>
<td>00</td>
<td>8.27±1.24</td>
<td>00</td>
<td>8.27±1.24</td>
</tr>
<tr>
<td>18 days</td>
<td>00</td>
<td>6.00±0.73</td>
<td>00</td>
<td>6.00±0.73</td>
</tr>
</tbody>
</table>

*Toluidine blue sections
Summary
The cellular emigration pattern has been studied in chicken by intradermal inoculation of E. coli. Heterophils and basophils were observed in insignificant numbers; however, monocytoid cells were highest in the early stages of chronic inflammation and decreased there after. This was followed by process of healing as evidenced by fibroplasia, angiogenesis and collagen formation.

References

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Occurrence of Polymastia in Sheep
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Abstract
Polymastia is a rare condition seen in Nilagiri sheep. Overall percentage of polymastia in Nilagiri sheep was 7.70 percent and the incidence of supernumerary teats were 2.20 per cent and 5.50 per cent for ewes with three and four teats, respectively.

Key words : Polymastia, Nilagiri sheep, Occurrence.

Nilagiri breed of sheep inhabiting the Nilgiris district of Soujh India is the only south Indian breed of sheep producing the apparel wool and the animal coat colour is white. Udder and teat characteristics are important determinants of milk yield and ease of milking in dairy animals (Rogers and Spencer 1991; De la Fuente et al. 1999). The present study deals with the occurrence of polymastia in Nilagiri sheep at the Nilgiris district, Tamil Nadu, India.

History and Observations
A total of 91 Nilagiri sheep were examined for polymastia in sheep farm at Karunkuzhimund, the Nilgiris district, Tamil Nadu. Biometry of all the teats was recorded to assess the length and circumference of teats and also distance between each teats. All the sheep were maintained under extensive system of management.

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