Effects of β–1,3/1,6-Glucan on Immune Responses in Broiler Chicks

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Measures of Preventing Diseases

- Immunoprophylaxis: Vaccines
- Antibiotics & Drugs
- Probiotics, Prebiotics & Synbiotics
- Immuno-Stimulants
Glucan

- **Polyglucose**
- $\alpha$-Linkage & $\beta$-Linkage
- $\beta$-Linkage : C(1-3), C(1-4), & C(1-6)
β-1,3/1,6-Glucan

- *Saccharomyces Cerevisiae*
- Structure Confirmed with NMR
- MW: 1500~50000 Da
Effects of $\beta$-1,3/1,6-Glucan on Macrophages \textit{in vitro}

- Two sources of macrophages
  - A transformed broiler splenic macrophage cell Line
  - Abdominal Exudate Macrophages from Broiler Chicks
- Exposed to various concentrations of $\beta$-glucan for 24h, then culture supernatants tested for nitrite levels
In vitro Stimulation of β-Glucan on Nitrite Production of MQ-NCSU Macrophage Cell Line (0.5×10^6/ml, 24h, 41°C, 5%CO₂)
In vitro Stimulation of β-Glucan on Nitrite Production of Broiler Macrophage (AE macrophages, $0.8 \times 10^6$/ml, 24h, 41C, 5%CO₂)
Nitrite production in culture

- β-1,3/1,6-Glucan by itself can induce nitric oxide synthase activity in macrophages after *in vitro* exposure.
In vitro Stimulation of β-Glucan on IL-6 Production of Broiler Macrophage (AE macrophages, $0.8 \times 10^6$/ml, 24h, 41°C, 5%CO$_2$)
IL-6 in culture

- β-1,3/1,6-Glucan exposure to AE macrophages in culture did not affect the IL-6 production *in vitro*
Feeding Trial

• One-day-old broiler chicks raised for 28 days
• Three Dietary Supplemental β-Glucan Levels:
  – In starter diets: 0, 20 and 40 mg/kg,
  – In grower diets: 0, 20 and 20 mg/kg
Experimental End Points

- Lymphoid Organ Wt. Relative to Body Weight (RW)
- Cell-Mediated Response (PHA-P toe web assay)
- Humoral Immune Responses (anti-SRBC antibody)
- Macrophage Functions: SRBC Phagocytosis, Nitrite and IL-6 Production
- CD4\(^+\)/CD8\(^+\) Incidence in the Intestinal Intra-epithelial Leukocytes (IELs)
Lymphoid Organ Weight of 2-Week-Old Broilers Fed Diet With β-Glucan

P=0.049
P=0.21
P=0.10
Lymphoid Organs

- Dietary $\beta$-Glucan treatment increased the relative weight of bursa and spleen of broiler chicks at the age of two weeks. However, this improvement was not observed at four weeks.
Effect of Dietary Supplemental β-Glucan on Response to PHA-P in Chick’s Toe web (18 days old)
Cutaneous Basophil Hypersensitivity

- β-Glucan treatment tended to maintain the PHA-P-mediated CBH response at relatively higher level for longer time
Effect of Dietary Supplemental \(\beta\)-Glucan on Total Antibody Response to SRBC in Broiler Chicks

\[\text{Total Antibody Titre, Log}_2\]

\[P = 0.012\]

\(\beta\)-Glucan

- 0
- 20-20
- 40-20
Antibody Response

• Only on day 4 after boost with SRBC, the total antibody titers were higher in β-Glucan-treated broilers than control, the difference is significant between control and the 40-20 mg/Kg (highest dosage) of β-Glucan supplemented dietary group (p <0.05)
Phagocytic Activity of AE Macrophages from Broilers Fed β-Glucan for 4 weeks

<table>
<thead>
<tr>
<th>β-Glucan, mg/kg</th>
<th>Phagocytosis, %</th>
<th>SRBC# per M.C.</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>34.02</td>
<td>1.8</td>
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<tr>
<td>20–20</td>
<td>44.61</td>
<td>2.7</td>
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<tr>
<td>40–20</td>
<td>41.04</td>
<td>2.8</td>
</tr>
</tbody>
</table>

P = 0.138

P = 0.0002
Phagocytic Ability of Macrophages

• Dietary supplemental β-Glucan tends to increase the phagocytic macrophages nearly 10%, even though the differences among treatments are not statistically significant (p>0.10).

• β-Glucan treatment resulted in significantly larger number of SRBC engulfed per phagocytic macrophage (p<0.01).
CD4⁺,CD8⁺ Lymphocyte Subsets in IELs from 2-Week-Old Broiler Chicks fed β-Glucan
IELs

• **β-Glucan may enhance intestinal mucosal immune responses**

  CD4+ & CD4+CD8+ double positive T cells, helper/inflammatory T cells, are supposed to respond to exogenous antigen in conjunction with class II MHC molecules

  CD8+ T cells, cytotoxic T cells, are supposed to respond to endogenous antigen in conjunction with class I MHC molecules

  IELs can produce IFN-γ and other cytokines. IFN-γ enhances surface expression of MHC molecules and augments IgA response
Conclusion

- β-Glucan can stimulate macrophages *in vitro* and dietary β-Glucan can increase the phagocytic activity of macrophages of birds.
- Dietary β-Glucan can enhance the humoral immuno-competence and intestinal mucosal immune response.
- β-Glucan can be used in diet as an immuno-modulator for broiler chicks.
Thanks!