Immunology of CBPP: current knowledge

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« Control of Emerging and Exotic Animal Diseases »
The pathogen

- *Mycoplasma mycoides subsp. mycoides (Mmm) “SC”*
- Class of *Mollicutes*
- Cell wall
- 2 genomes fully sequenced
- PG1 and Gladysdale
- 1,211 kb
- NGS based phylogeny
- Molecular dating (Dupuy et al. 2012)
The disease

- contagious bovine pleuropneumonia (CBPP)
- Africa
- lymphatics / lungs

- chronic CBPP
- sequestrum
- long term persistance
- and excretion?

- acute CBPP (15-30%)
- massive inflammation of lungs
- hepatisation
- pleurisy
Need for improved Vaccines

Currently: Live, empirically attenuated strains injected sub-cutaneously

Advantages
- Relatively low production costs
- Very long conservation at -20°C once freeze-dried
- Easy administration (sub-cutaneous)
- T1sr: completely safe
- Transient sero-conversion (allows detection of outbreaks)
- Repeated vaccinations result in good protection

Drawbacks
- Thermolability (freeze-dried or reconstituted)
- Freeze-drying needs industrial skill
- T1/44: some residual virulence (primo-vaccination)
- Lack of sero-conversion does not allow sero-monitoring of vaccination campaigns
- A single administration does not yield good protection
- Protection is short-lived (T1sr: 6 months, T1/44: one year)
- Eradication cannot be achieved with vaccination alone

Willems reaction
- With T1/44 (not T1sr)
- In animals vaccinated for the first time
- Appears 10-20 days post vaccination
- 0 to 5% of vaccinees
# Immunology of CBPP: current knowledge

## 1st part

<table>
<thead>
<tr>
<th></th>
<th>Innate immunity</th>
<th>Acquired immunity</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Primary infection</strong></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td><strong>After recovery</strong></td>
<td></td>
<td>+</td>
</tr>
<tr>
<td><strong>After vaccination</strong></td>
<td></td>
<td>+</td>
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</tbody>
</table>

**Summary of Knowledge and gaps**

## 2nd part

**opportunities from the CBPP BEN1 vaccine project**
Immunology of CBPP: current knowledge

Innate immunity

• during primary infection (injection subcut/intubetion/contact)

  Lesions in lungs and locally (subcut) are typical of inflammation. Less severe
  when injection at the tip of the tail → importance of the lymphatic system

  presence of myeloid cells (iL24) in lesions.

  TNF-α is produced in alveolar macrophages in response to both pathogenic
  and non pathogenic MmmSC (in vitro)
  (Jungi T et al. Microbial pathogenesis 1996)

  Archetypal inflammatory cytokines (pro and anti) are detected early in the plasma
  and at higher levels in acute cbpp (n=4). No effect of CD4+T cells depletion.
  (Sacchini F et al. BMC Vet Res 2012)

Summary:

➢ No endotoxins/ Mechanisms of inflammation (chimiokines)?
➢ Very little data on MmmSC-host cells interactions/ role of epithelial lung cells?
➢ Which cell types are producing proinflammatory cytokines?
Humoral immunity

- during primary infection (contact challenge/intubation)

  - Lesions size correlates with *Mmm*SC-specific IgM titers in serum (CFT, agglutination)

  - Detection of *Mmm*SC-specific IgA in serum and bronchoalveolar fluids (ELISA) of animals with less severe disease (n=5) / detectable for only 1-3 months in serum (Niang M et al. Vet Res. 2006)


  - higher early anti-*Mmm*SC IgG1and G2 titers (ELISA) in severe cbpp after intubation (n=4) (Sacchini F et al. BMC vet res 2012)

Summary;

- Possible role of immune complexes (IgM, IgG1, IgG2) in the pathology and IgA in protection

- Proteins targetted? → potential of luminex approach with individual surface proteins
Humoral immunity

• during primary infection (contact challenge/intubation)

● during vaccination (T144/T1sr)

- Weak antibody titers (CFT, cELISA) and most animals are negative after 3 months. No correlation with protection after contact challenge

- Strong IgG titers against 4 out of 65 recombinant surface proteins (luminex) with LppQ as potential indicator of protection after contact challenge (n=5)

Summary:

➢ Potential of the luminex methodology to identify markers of vaccine success before challenge
Cell mediated immunity (CMI)

- during primary infection (contact/intubation challenge) / peripheral blood

  - Sustained *MmmSC*-specific recall activation of CD4+T lymphocytes and IFN-\(\gamma\) production is associated with a better control of cbpp
    *(Dedieu L et al. Vet Immunol Immunopathol 2005)*

  - No correlation between *MmmSC*-specific recall IFN-\(\gamma\) production and protection after intubation. No build up of T-lymphocytes in lung lesions but presence of myeloid cells.

  - Depletion studies indicate that removal of CD4+T lymphocytes has no incidence on disease progression *(Sacchini F et al. Vet Res 2011)*

Summary:

- No role for CD4 in primary infection
Cell mediated immunity (CMI)

- during primary infection (contact/intubation challenge) / peripheral blood
- after recovery (fibrotic scars) / draining lymph nodes → memory + protection

→ *MmmSC*-specific recall activation of CD4+ T lymphocytes but no proliferation (n=5) (*Dedieu L et al. Vet Res 2006*)

→ *MmmSC*-specific recall proliferation of B and CD4+ T lymphocytes / strictly CD4-dependent / CD4 express memory markers, produce IFN-\(\gamma\) but no IL-4 (Th1 or Th17?) (*Totté et al. Vet Res 2008*)

→ *MmmSC*-specific memory CD4 comprise Tem (short lived) and Tcm (long lived) (*Totté et al. Dev Comp Immunol 2010*)

Summary:

- Strong CD4 memory response after recovery → role in protection against secondary challenge?
- Potential of CD4+Tcm as markers of protection as in bovine tuberculosis
Effector memory (Tem) vs central memory (Tcm)

**Tem**
- blood-tissues-lymph circulation (ccr5, cxcl3)
- low proliferation
- strong IFN-γ response
- Months after infection/immunisation

**Tcm**
- blood-lymph circulation (cd62l, ccr7)
- high proliferation
- low IFN-γ response
- years after infection/immunisation
- differentiate into T effectors upon Ag encounter

Improved vaccines → Tem + Tcm

- local response to challenge but short-lived
- Long-lived pool of effectors

Strong challenge → long term immunity
Cell mediated immunity (CMI)

- during primary infection (contact/intubation challenge) / peripheral blood
- after recovery (fibrotic scars) / draining lymph nodes

- after vaccination (T144/T1sr) / peripheral blood

- no recall activation of lymphocytes in cattle vaccinated 2 months previously in the tail tip with a single dose of the T1 vaccine

- \(MmmSC\)-specific recall IFN-\(\gamma\) and proliferation of CD4+Tem and Tcm but only after 3 shots of T1 vaccine \((n=5)\) / lower Tcm response in comparison to animals that recovered from CBPP
  \[(\text{Totté P et al. PloS One 2013})\]

➢ Summary:

➢ Low T-cell immunogenicity of T1 vaccines

➢ Immunoinhibitory mechanisms?
Immunology of CBPP: opportunities from the CBPP BEN1 vaccine project

- confirm in larger animal groups data obtained previously
  - IgG1, IgG2 and IgA responses against individual or groups of specific antigens as indicators of protection/pathology (ILRI)
  - Poor T-cell immunogenicity induced by live vaccines (ILRI)
  - Correlation between higher production of inflammatory cytokines early after challenge and disease severity (?)
Immunology of CBPP: opportunities from the CBPP BEN1 vaccine project

- Improve our current understanding of CBPP immunology

- Characterize global immune protective mechanisms induced by attenuated CBPP vaccines (CIRAD+ILRI)

- Involvement of various T-cells sub-types (Th orientation) in vaccinated animals undergoing challenge (ILRI+CIRAD)

- Characterize global immune responses and cell types involved in the pathology (HVRI+CIRAD)

- Characterize the role of EBL in innate immune responses to MmmSC infection (HVRI)
Immuneology of CBPP: opportunities from the CBPP BEN1 vaccine project

- definition of markers that can predict vaccine efficacy after vaccination but before challenge (CIRAD+ILRI)

Lessons from the bovine tuberculosis model (20 years of research on CMI in cattle and 10 years on correlates of protection)

- whole blood and T-cell based assays (recall IFN-γ response) are good markers of protection at group level but NOT at individual level

- IFN-γ cultured ELISPOT (Tcm) and intracellular INF-γ labeling in stimulated CD4 are good correlates of protection at individual level (R²=0.79 to 0.83) but complex and time consuming (Hope JC et al. Clin Vaccine Immunol 2011)

- whole bovine genome transcriptomics has identified a 3-genes signature (il-22, ifn-γ, mt3) that can predict protection following vaccination (14 weeks) and before challenge confirmed by rt-qPCR in independent experiment (n=16) (Bhuju S et al. PLoS Pathog. 2012)