INFECTION DIAGNOSIS IN SYSTEMIC INFLAMMATION BY INNATE IMMUNE RECEPTOR EXPRESSION PATTERN

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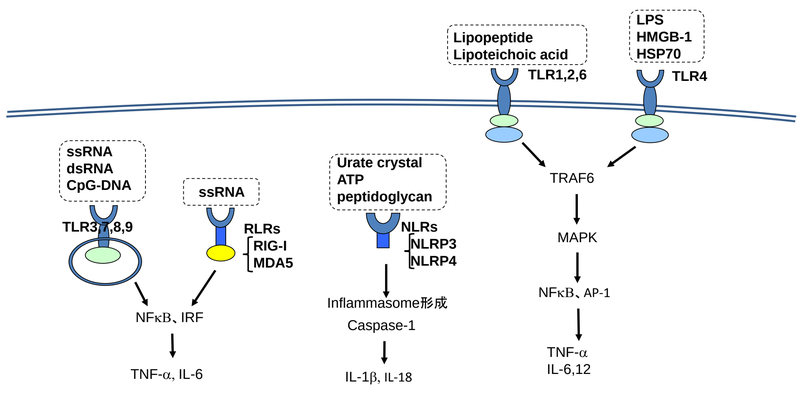
【Abstract】

Introduction: It is difficult to diagnose infection by single biomarker in patients who are under condition of systemic inflammation. We hypothesized that expression pattern of innate immune receptors may distinguish infection from systemic inflammation of uncertain etiology.
Methods: To compare infectious inflammation and sterile inflammation, we employed cecal ligation and puncture (CLP) and 20% full thickness burn injury (Burn) model. C57BL/6 mice underwent sham, CLP, or Burn. 24 hours later, mice were sacrificed, and total RNA was extracted from whole blood. Using quantitative real-time PCR, we investigated gene expression of innate immune receptors including TLR2, TLR4, TLR9, NLRP3 and RIG-I. To evaluate all the gene expression together as patterns, each value was plotted on the radar chart and the area was calculated. To compare gene expression patters as graphic characters, area A / (B+C+D+E) was defined as bacterial infection index (BI) and evaluated.
Results: Gene expression of TLR2, TLR4 and NLRP3 was significantly increased in both CLP and Burn compared to sham ($p<0.05$). Gene expression of TLR9 was significantly decreased in CLP compared to both sham and Burn ($p<0.05$). RIG-I gene expression did not show any difference (Fig.1). In the radar chart, each group showed distinctive gene expression patterns (Fig.2a). BI in CLP was significantly higher than sham and Burn ($p<0.05$, sham: min=0.19, max=0.25, mean=0.23, CLP: min=1.24, max=3.01, mean=2.26, Burn: min=0.58, max=0.77, mean=0.67), and BI higher than 1.0 distinguished infection clearly from the other groups (Fig.2b).
Conclusion: Gene expression profile of innate immune receptors distinguishes infection from sterile systemic inflammation. BI can assess multiple factors together, and will be convincing marker to diagnose infection.

【Background】

It is difficult to diagnose infection by single biomarker. Even if the cause of inflammation is different, same signal molecules or cytokines are expressed.

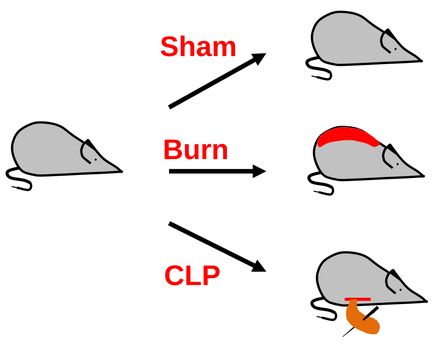


【Hypothesis】

Expression pattern of innate immune receptors may distinguish infection from systemic inflammation of uncertain etiology.

【Methods】

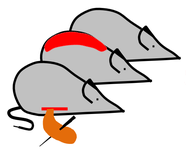
Animal model



Sterile Inflammation model:
25%TBSA Burn

Infection model:
CLP(cecal ligation and puncture)

Quantitative RT-PCR



Sham, Burn or CLP

24h

Blood Sampling

mRNA extraction
Quantitative RT-PCR

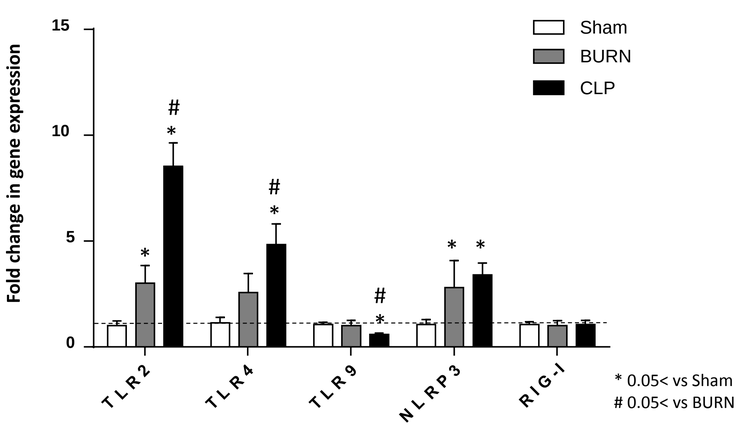
- **Target gene: Tlr2, Tlr4, Tlr9, Nlrp3, Ddx58**
- **Reference Gene: Rps18**

➢ To evaluate all the gene expression together as patterns, each value was plotted on the radar chart and the area was calculated.

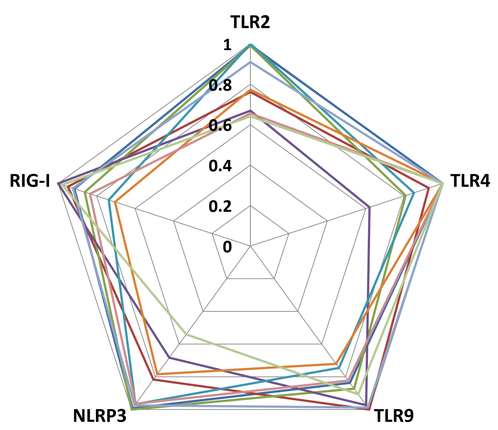
➢ To compare gene expression patters as graphic characters, area A / (B+C+D+E) was defined as bacterial infection index (BI) and evaluated.

【Results】

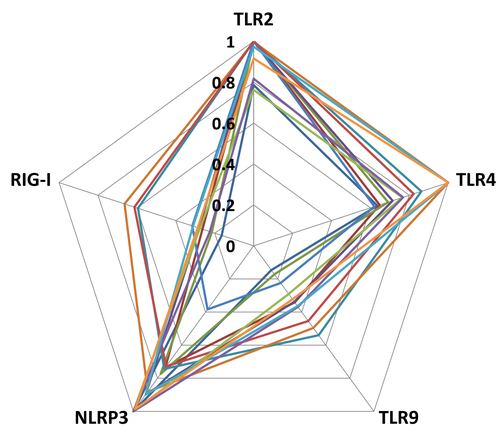
mRNA expression



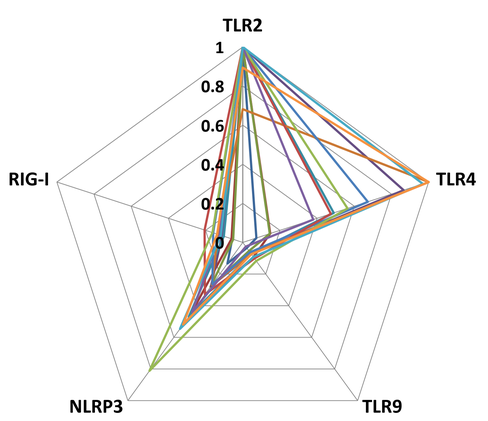
Sham



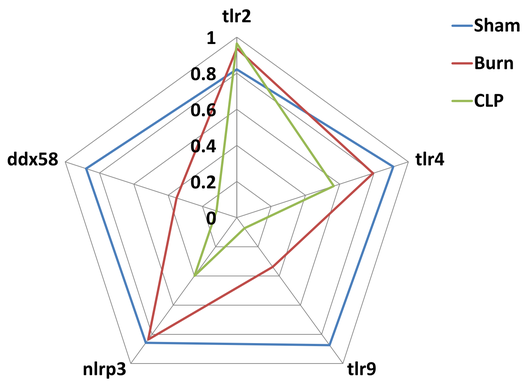
Burn

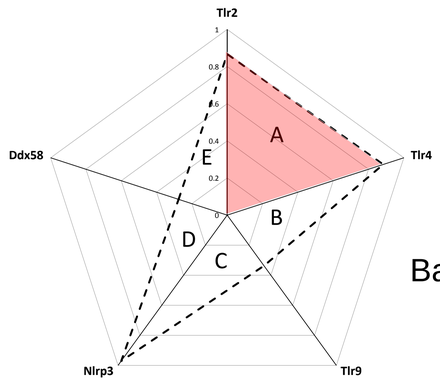


CLP

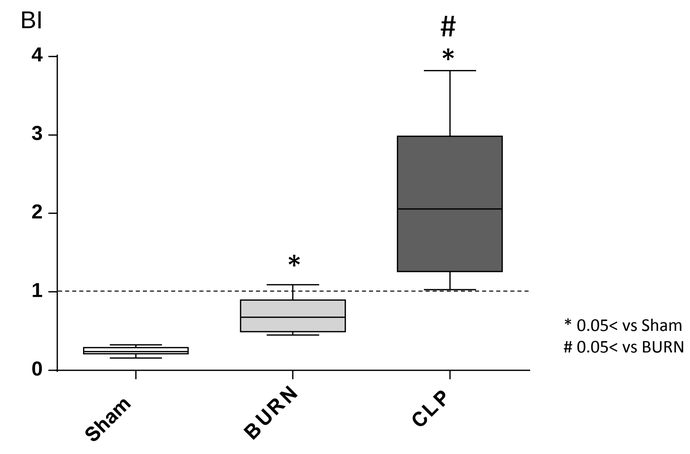


Bacterial Infection Index





Bacterial Infection Index：BI
=Area（A / B+C+D+E）



【Future Direction】

- Time course study
- Activation of downstream signaling pathway

【Conclusion】

- Gene expression profile of innate immune receptors distinguishes infection from sterile systemic inflammation.
- BI can assess multiple factors together, and will be convincing marker to diagnose infection.