Innate immune activation in AML12 hepatocyte cells is NFkB dependent
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INTRODUCTION
The hepatic innate immune response plays a central role in protection against sepsis.

The innate immune response is well understood to be activated by LPS (lipopolysaccharide) in macrophages and neutrophils.

NFkB is the primary regulator of the innate immune response, activating downstream inflammatory genes.

We hypothesize that hepatocytes can mount an immune response through the activation of NFkB and its downstream target genes.

METHODS
- **Cell culture model**: Cultured AML12 hepatocyte cells
- **Cell culture treatments**: Cells were exposed to the following treatments:
  - LPS dose 15, 30, 60 min: 100ng/ml; BAY dose, 30min: 5, 10, 20uM; IL-1B dose 10ng/ml; and/or IL-6 dose 1, 5hr: 10, 20ng/ml
- **Outcome measures**: mRNA expression was evaluated by qPCR for cxcl10, icam, tnf, ikba, il6
- **Protein expression of cellular p65, p50, ikbα, ikbβ** was assessed using Western Blot
- **Statistical analysis** was performed by 2-way ANOVA with post hoc analysis using GraphPad prism and significance defined as p<0.05

RESULTS

**Fig 1a**: LPS induces NFkB activation in AML12 cells via subunit nuclear translocation and IKB degradation

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<thead>
<tr>
<th>LPS 100ng/ml</th>
<th>Nuclear</th>
<th>Cytoplasmic</th>
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<tr>
<td>Cont</td>
<td>15 min</td>
<td>30 min 60 min</td>
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<tr>
<td>p65</td>
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**Fig 1b**: LPS induces NFkB activation in AML12 cells via cytokine gene expression

**Fig 2**: NFkB inhibitor, BAY, results in decrease of inflammatory genes expression

CONCLUSION

The AML12 hepatocyte cell line responds to inflammatory exposures by initiating an innate immune response characterized by activation of NFkB

These results suggest that treatments directly targeting the hepatocyte innate immune response may be a novel therapeutic strategy

Future direction includes assessing the BAY effect on NFkB nuclear subunit translocation and IKB degradation and further describe the importance of these pathways in liver disease