

Mint oils:

in vitro anti-inflammatory effects tested in porcine alveolar macrophages

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OVERVIEW



- Introduction
- Research objective and methods
- Results
- Conclusions





INTRODUCTION



PORCINE ALVEOLAR MACROPHAGES (PAMs)

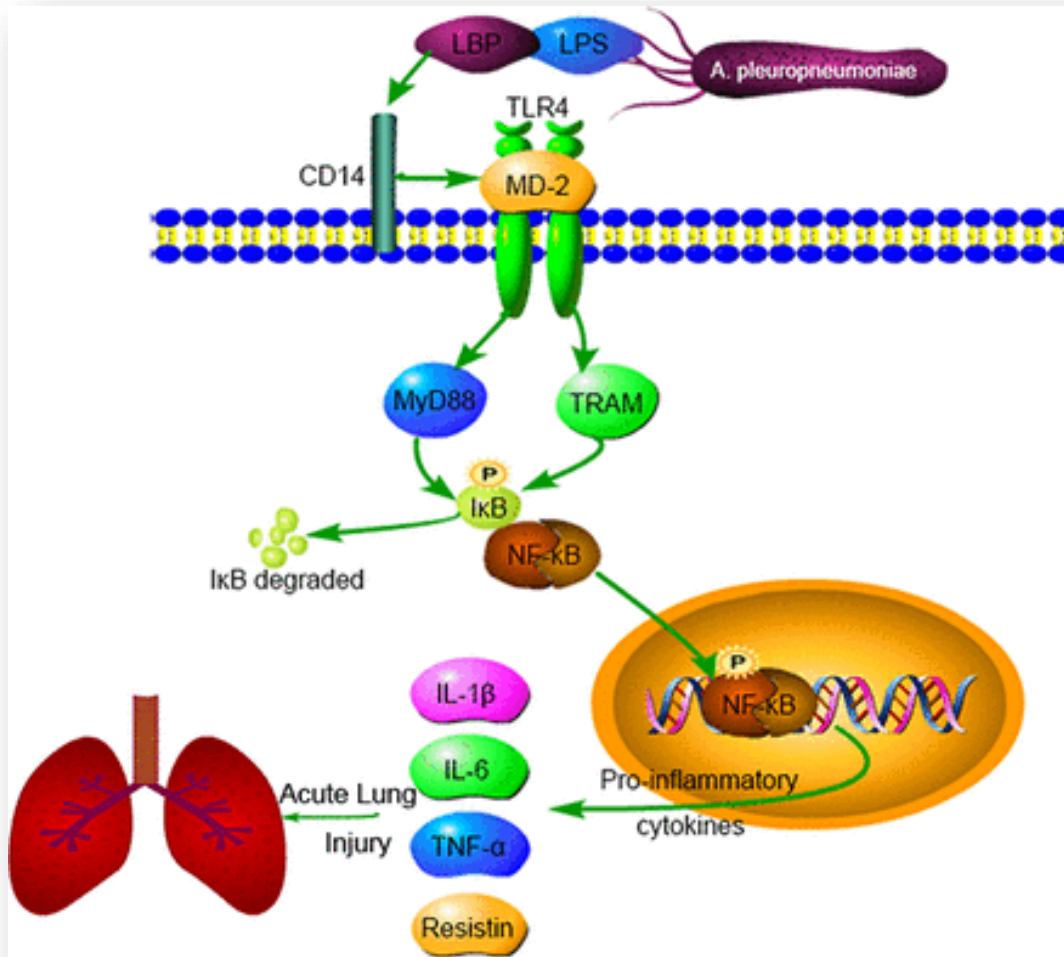


Fig 1. LPS induces the production of TNF- α in PAMs cells. (Li et al. 2018).



INTRODUCTION



ESSENTIAL MINT OILS

- *Mentha* (mint) is a genus of an aromatic herb and pepper mint and spearmint are among the most important crops in EOs production owning the abundant content of **phenolic** compounds.

(Wu et al. 2019)



Fig 2. Peppermint oil (*Mentha piperita* L.).



Fig 3. Spearmint oil (*Mentha spicata* L.).



OBJECTIVE



The research objective of the study was to measure the *in vitro* anti-inflammatory effects of peppermint oil and spearmint oil with porcine alveolar macrophages as host immune responses.



PAM cell

Peppermint oil
Spearmint oil

+/-
LPS

Concentration
of TNF- α



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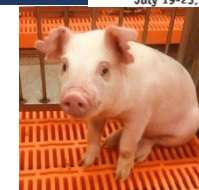
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EXPERIMENTAL DESIGN AND TREATMENT

- **RCBD** with dose and LPS as fixed effects and pig as random effects (n=12).
- The PAM cells were treated with pepper and spearmint oils:
2 (doses of LPS – 0 or 1 $\mu\text{g/mL}$) \times 5 (doses of mint oil – 0, 25, 50, 100, 200 $\mu\text{g/mL}$).





SAMPLE COLLECTION AND ANALYSIS

- Macrophages were harvested from the bronchial lavage of **6 pigs** at **6 weeks** of age and seeded into 24-well plates with at 10^6 cells/mL.
- The concentration of $\text{TNF-}\alpha$ was performed by ELISA assay into 96-weel plates.
- Cell viability was tested by the MTT assay.
- Data were analyzed by the MIXED of SAS 9.4.

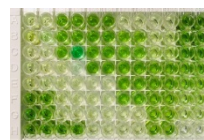
The protocol for this study was approved by the Institutional Animal Care and Use Committee at the University of California, Davis.

	1	2	3	4	5
A	0	25	50	100	200
B	NO LPS	NO LPS	NO LPS	NO LPS	NO LPS
C	<u>0</u>	<u>25</u>	<u>50</u>	<u>100</u>	<u>200</u>
D	<u>LPS</u>	<u>LPS</u>	<u>LPS</u>	<u>LPS</u>	<u>LPS</u>

Fig 4. The graphical presentation of PAM cells seeded into 24-well plate

	1	2	3	4	5	6	7	8	9	10	11	12
A	0	<u>100</u>	<u>25</u>	200	50	2000						
B	25	<u>200</u>	<u>50</u>	<u>0</u>	100	1000						
C	50	0	<u>100</u>	<u>25</u>	200	500						
D	100	25	<u>200</u>	<u>50</u>	<u>0</u>	250						
E	200	50	0	<u>100</u>	<u>25</u>	125						
F	<u>0</u>	100	25	<u>200</u>	<u>50</u>	62.5						
G	<u>25</u>	200	50	0	<u>100</u>	31.3						
H	<u>50</u>	<u>0</u>	100	25	<u>200</u>	0						

Legend:



Peppermint NO LPS	<u>Peppermint LPS</u>
Spearmint NO LPS	<u>Spearmint LPS</u>

Fig 5. The graphical presentation of MOs seeded into 96-weel ELISA plates.



RESULTS



PEPPERMINT OIL

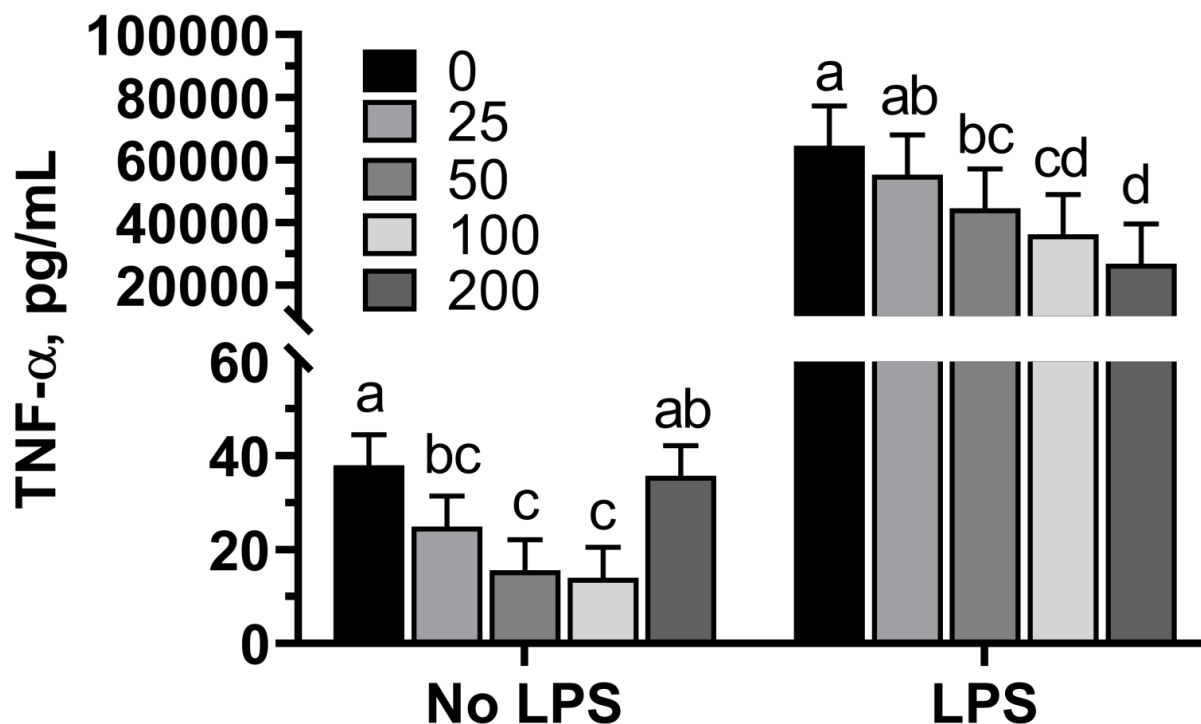


Fig 6. The concentration of TNF- α secreted by PAM treated with peppermint oil influence the production of tumor necrosis factor- α (TNF- α) in the absence or presence (1 μ g/mL) of lipopolysaccharide (LPS). Cells were incubated with various peppermint concentrations (0, 25, 50, 100, 200 μ g/mL) for 24h. All dose were not toxic for the cells according to MTT tests. The results were means of values from 6 pigs.



RESULTS



SPEARMINT OIL

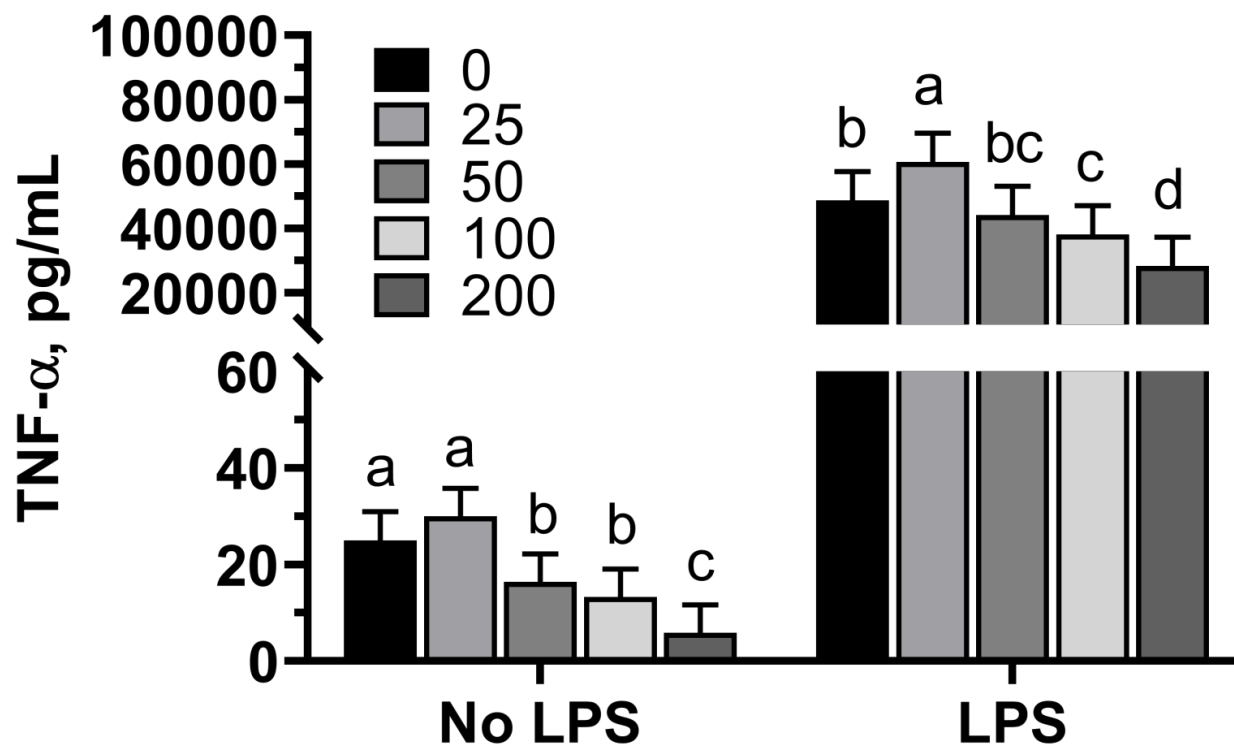


Fig 7. The concentration of TNF- α secreted by PAM treated with spearmint oil influence the production of tumor necrosis factor- α (TNF- α) in the absence or presence (1 μ g/mL) of lipopolysaccharide (LPS). Cells were incubated with various spearmint concentrations (0, 25, 50, 100, 200 μ g/mL) for 24h. All dose were not toxic for the cells according to MTT tests. The results were means of values from 6 pigs.





CONCLUSION



The *in vitro* experiment on PAM with mint oils indicated that:

- Mint oils **dose-dependently reduce** the secretion of TNF- α by PAMs challenged with LPS (1 $\mu\text{g/mL}$).
- Thus, peppermint and spearmint oils had **anti-inflammatory** activities *in vitro*.
- *In vivo* animal trials will be conducted to evaluate their impacts on animal health and performance.



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THANK YOU FOR YOUR ATTENTION!

