INVESTIGATION OF THE ANTI-INFLAMMATORY EFFECT OF LIQUIDAMBAR *ORIENTALIS* LEAF EXTRACT ON RAW 264.7 MACROPHAGE CELLS

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OUTLINES

- Lquidambar Orientalis
- Aim of Research
- Method
- Results
- Conclusion

Altingiaceae Familia

Lquidambar *Orientalis* (Sweetgum)

- L. *Orientalis*, known as the Anatolian Sweetgum Tree, is an endemic species.

- Mainly, it is distributed in the southwestern regions of Turkiye, especially in Köycegiz, Fethiye and Marmaris.

- It has been used for centuries to treat diseases such as ulcers, gastritis, dermatitis and eczema by local people.

Aim Of The Research

1

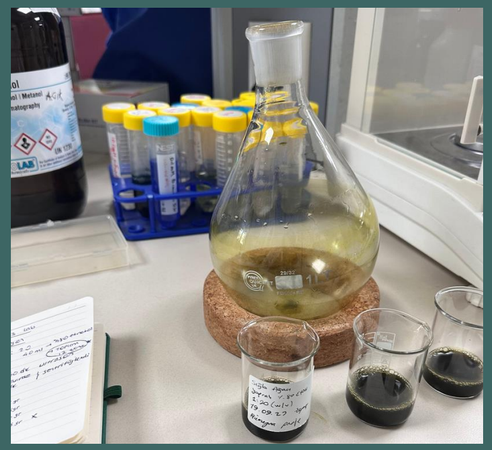
The demonstration of anti-inflammatory effect of L. *Orientalis* leaf extract in vitro

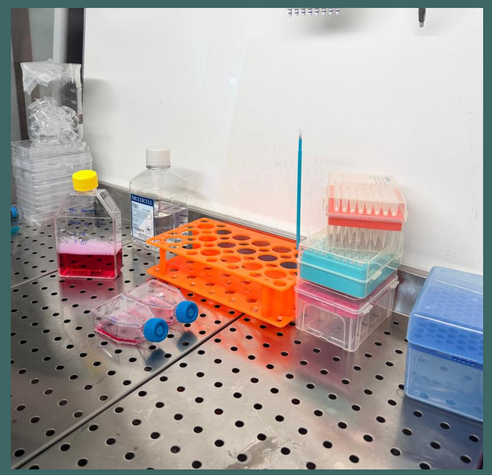
2

Investigation of the potential mechanism underlying its anti-inflammatory activity

3

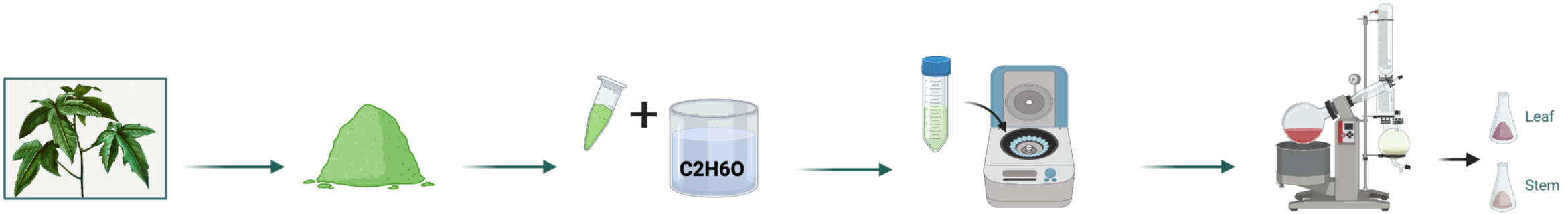
Comparison of L. *Orientalis* extract and methotrexate Which is used commonly for treatment





METHOD

Figure 1: Preparation of L. *Orientalis* Leaf and Stem Extract



Leaf and stems collection of L. *Orientalis*

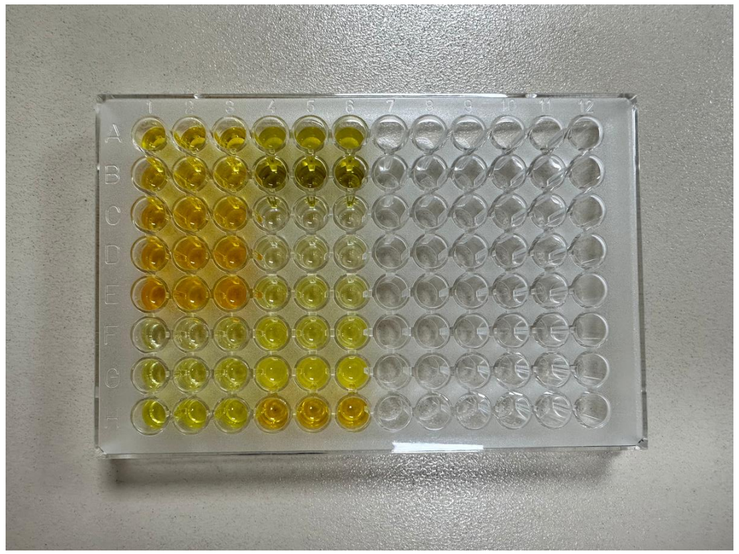
Drying and pulverizing leaves and stems

Leaf and stem samples mixed with 80% ethanol at the ratio of 1:20 (w/v)

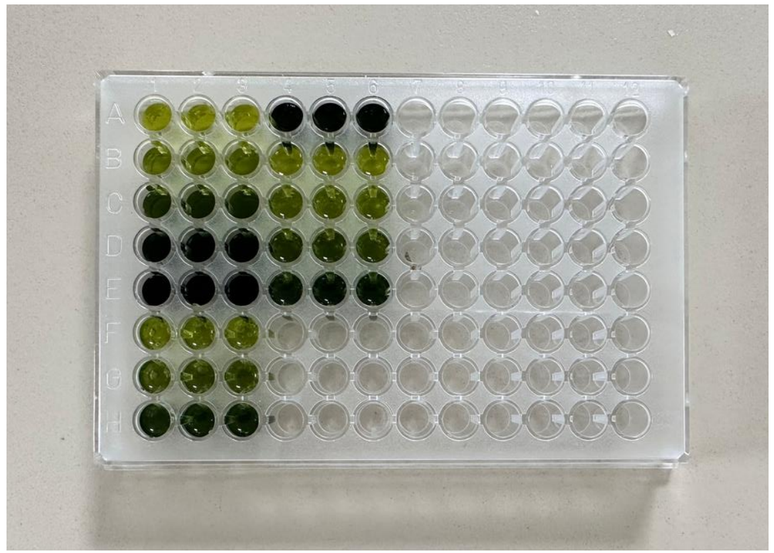
Ethanol extract of samples was centrifuged after 60 minutes of ultrasound at 400 W and 45°C

All supernatants were concentrated in the lyophilizer and stored as powder extract.

2. Determination of Total Flavonoid and Phenolic Contents



Phenolic Content



Flavonoid Content

- In different concentrations, leaf and stem extract solutions phenolic and flavonoid content were measured by spectrophotometer.

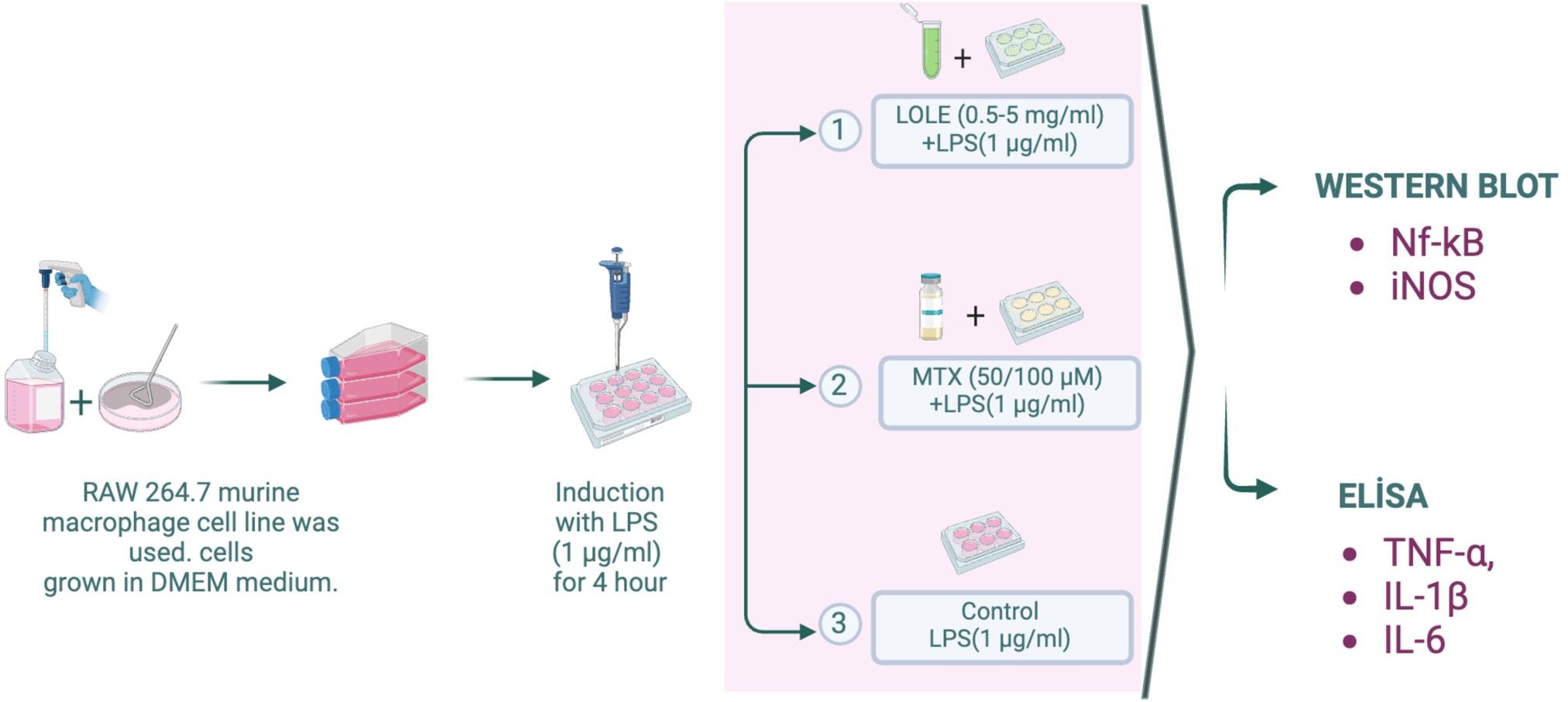


Figure 2: Preparation of Cell Culture and measurement of inflammatory markers by Western Blot and ELISA

*Lipopolysaccharide (LPS), L. orientalis leaf extract (LOLE), Methotrexate (MTX), Nitric Oxide Synthase (iNOS), Nuclear Factor-kappa B (NF-κB), Tumor Necrosis Faktor alfa (TNF-*α), Interleukin-1β (IL-1β)

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RESULTS

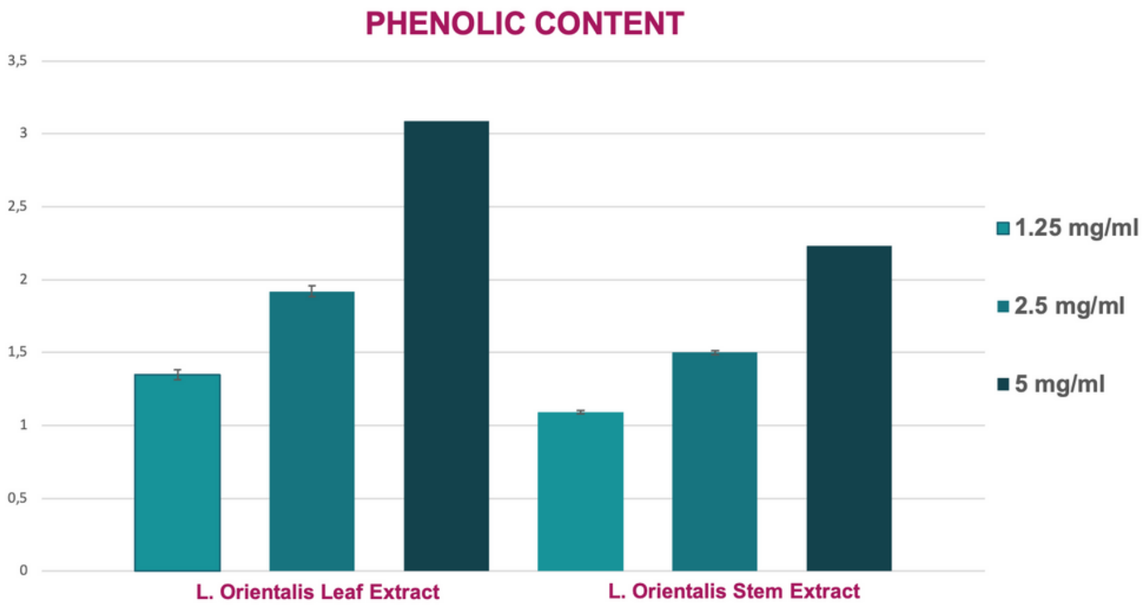


Figure 3: Dose-dependent comparison of total phenolic levels of L. *Orientalis* leaf and stem extract

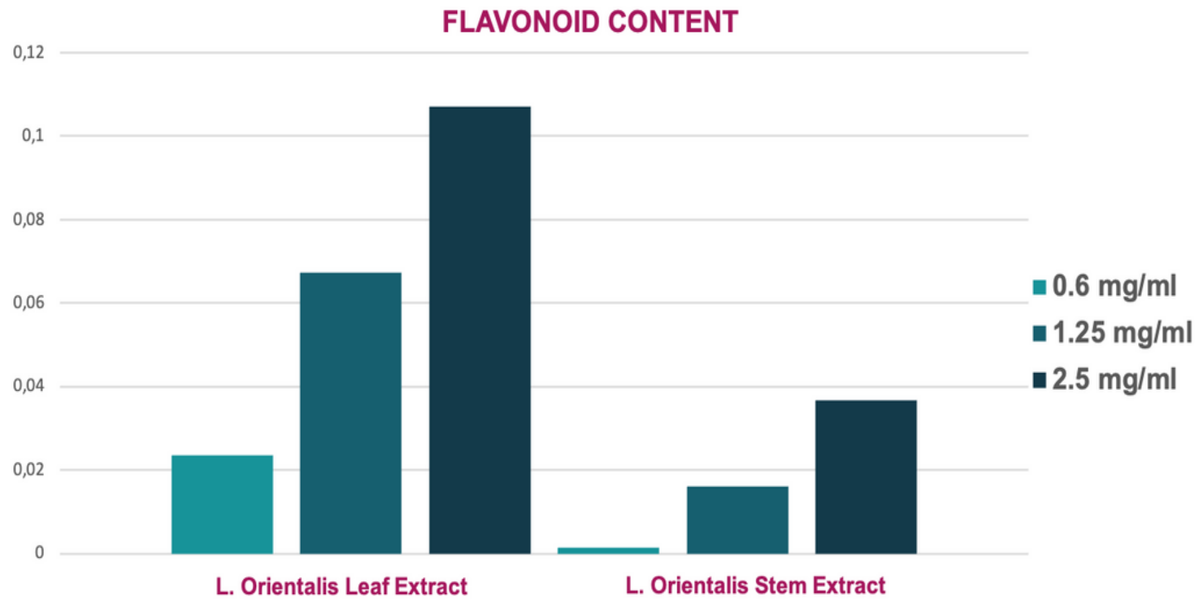


Figure 4: Dose-dependent comparison of total flavonoid levels of L. *Orientalis* leaf and stem extract

ELISA Results

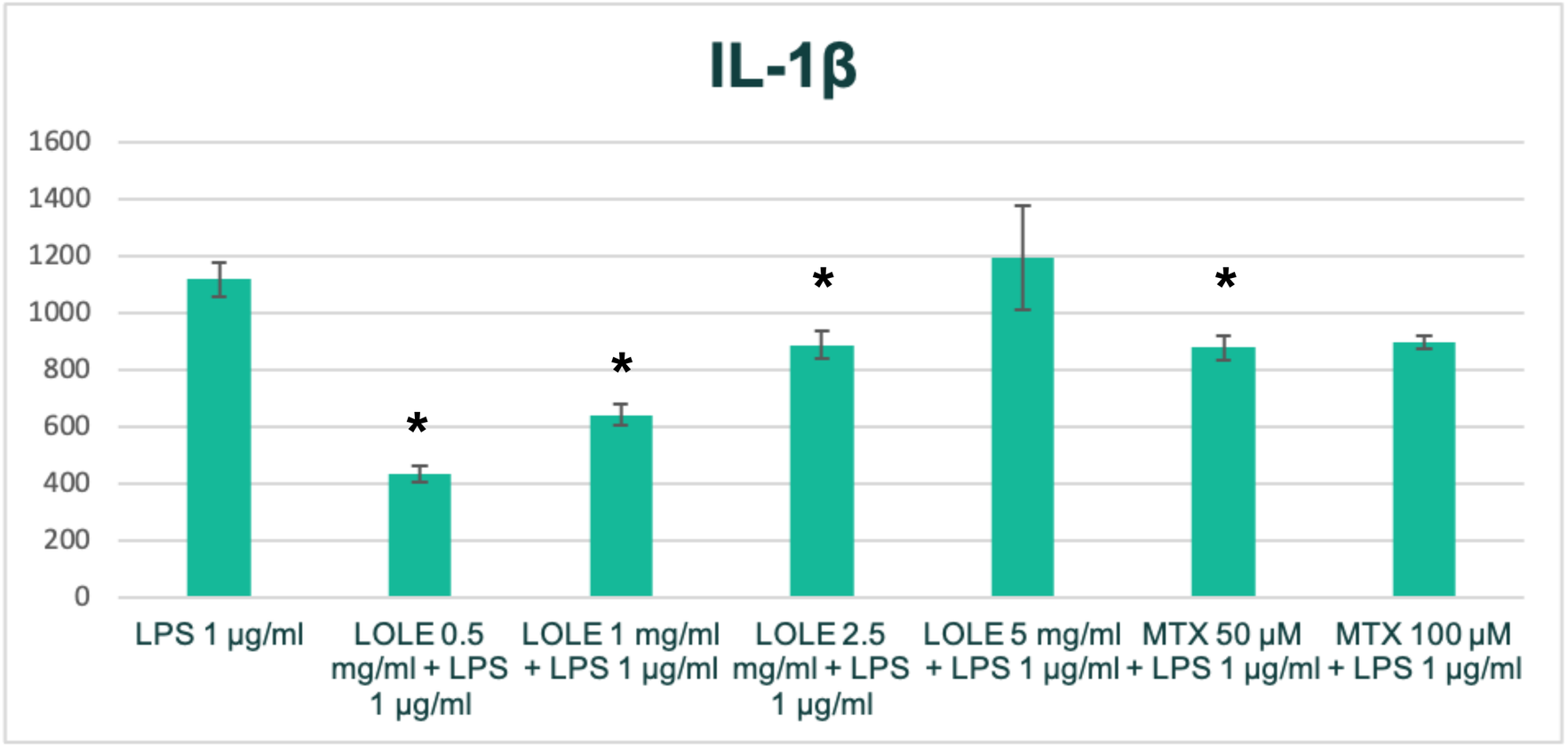
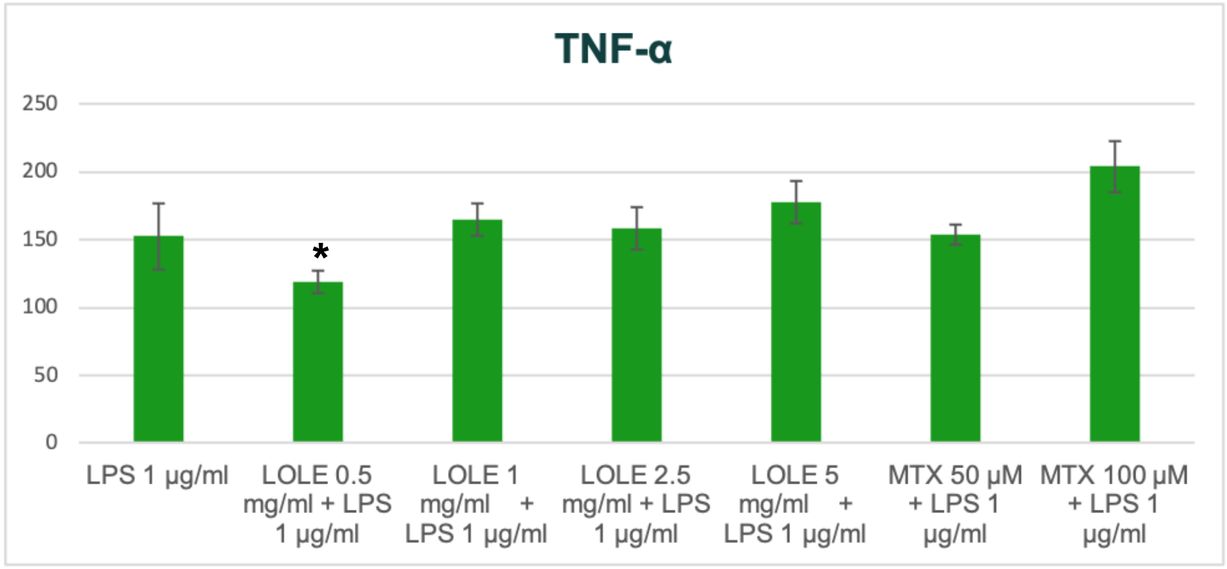


Figure 5: Demonstration of the changes in IL-1β levels within LPS-induced macrophage cells in response to the addition of LOLE or MTX at different concentration ($p<0.05$)

(Lipopolysaccharide (LPS), L. orientalis leaf extract (LOLE), Methotrexate (MTX))



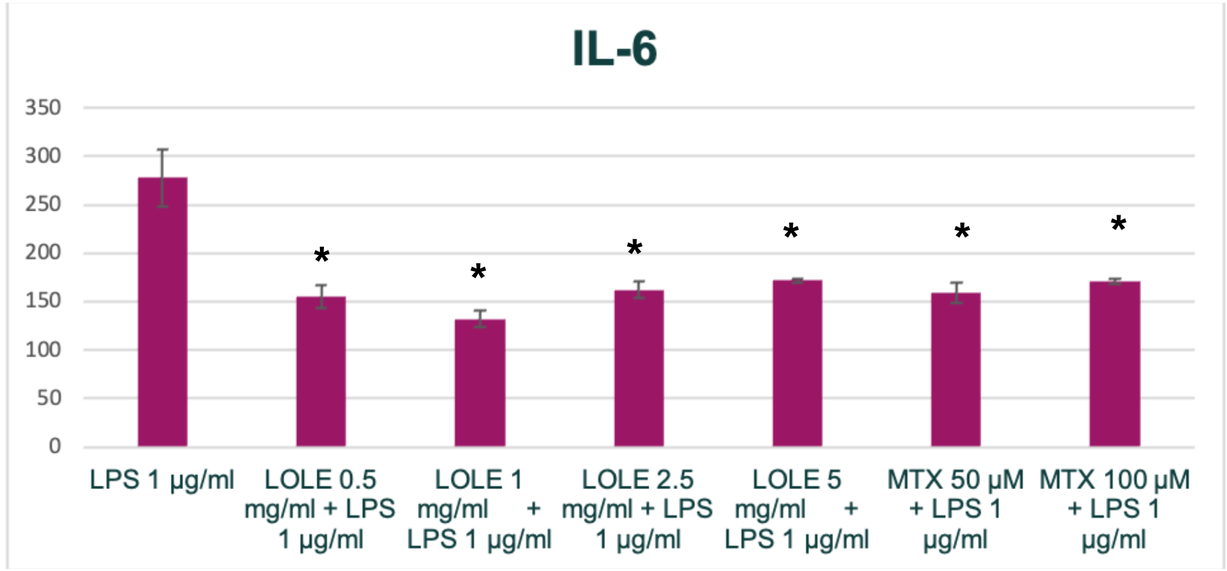
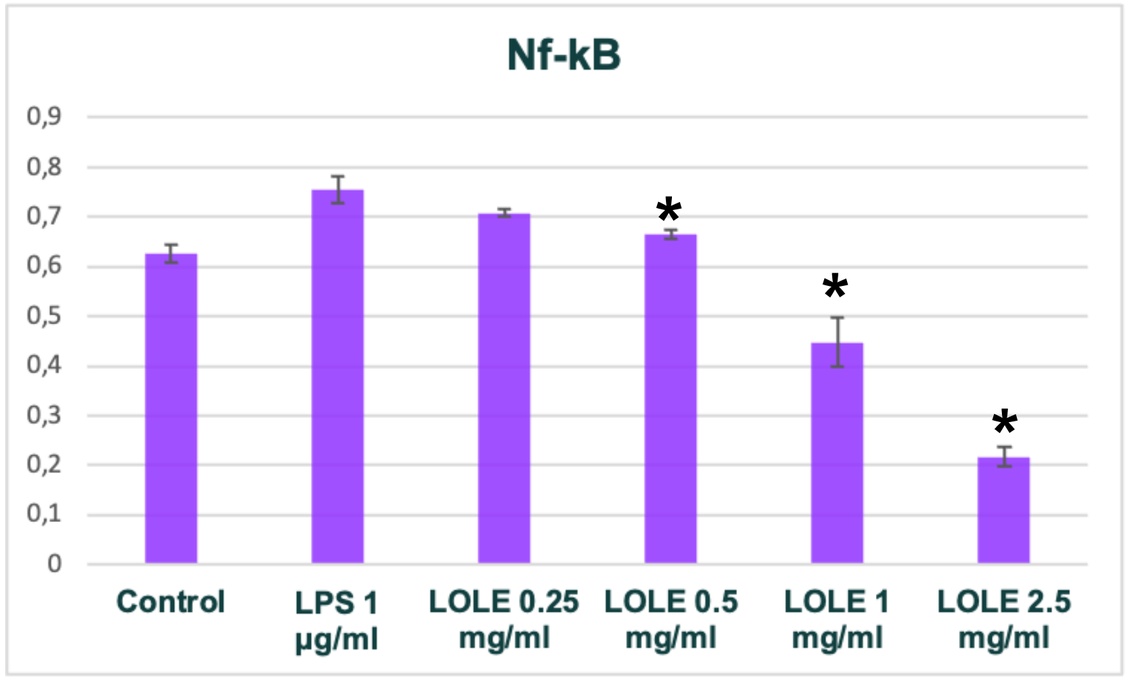
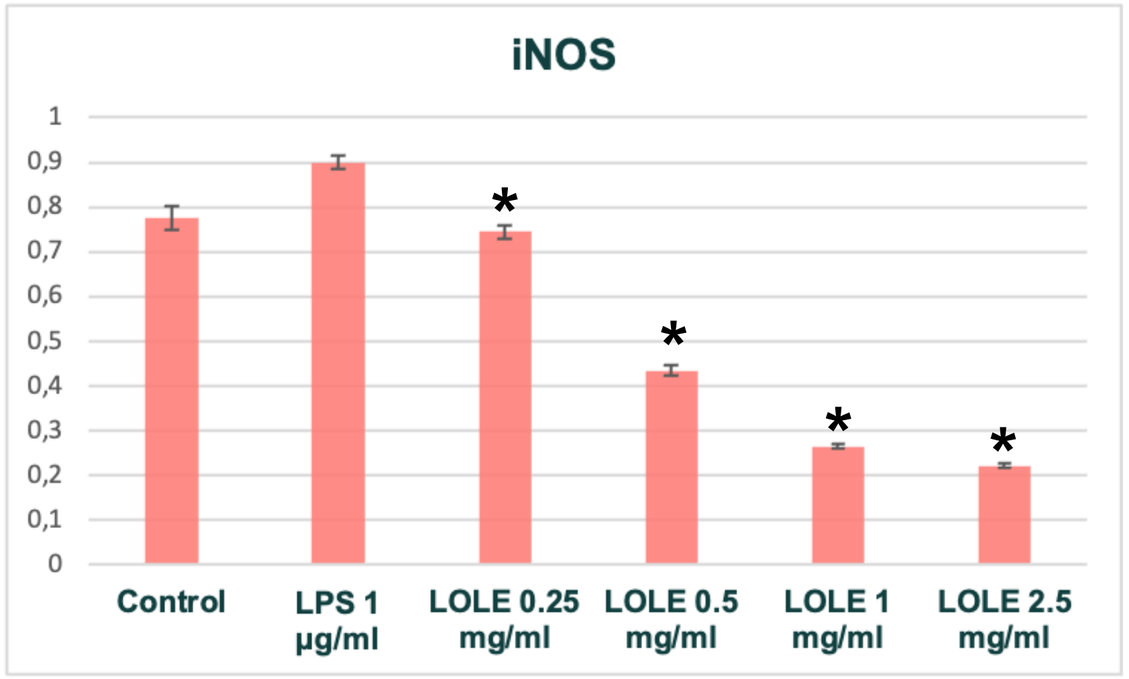


Figure 6 : Demonstration of the changes in IL-6 and TNF-α levels within LPS-induced macrophage cells in response to the addition of LOLE or MTX at different concentration. ($p<0.05$)

(Lipopolysaccharide (LPS), L. orientalis leaf extract (LOLE), Methotrexate (MTX), Tumor Necrosis Factor alfa (TNF-α))

Western Blot Result



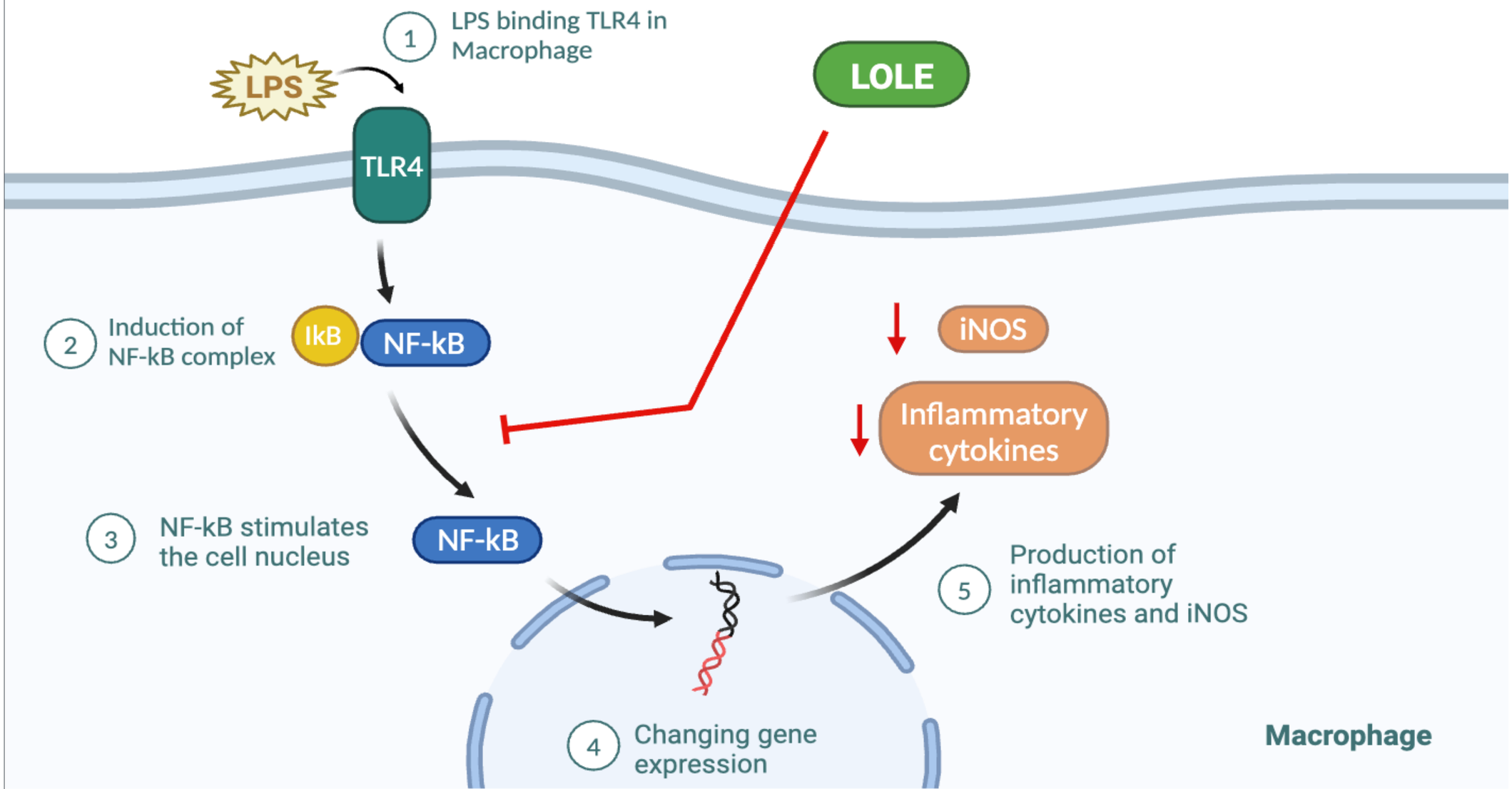


Western Blot results demonstrated that LOLE suppressed the expression of inflammation-related proteins iNOS and NF-κB in LPS induced macrophage cells, in a dose-dependent manner. ($p<0,05$)

(Nuclear Factor Kappa B (NF-kB), Inducuble Nitric Oxide Synthase (iNOS))

LPS İnduced Macrophage cell

Potential Mechanism Of LOLE



CONCLUSION

1- LOLE decreased inflammatory cytokines I**L-1β, IL-6, and TNF-α** through its anti-inflammatory effect.

2- *In vitro* studies have demonstrated that LOLE suppress IL-1β levels more than MTX.

3- One of the potential mechanisms underlying the anti-inflammatory effect of LOLE could be the modulation of **NF-kB signaling pathways.**

CONCLUSION

4- Since *in vitro* studies have limitations, the results should be supported by *in vivo* experiments.

5- Further analysis are required to explore other mechanisms underlying the anti-inflammatory effect.

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First Lab Day

19.09.2023

Thank you For contributions

- Associate Professor Dr. Ozge Pasin
- Research Assistant Ezgi Durmus

THANK YOU

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