

Introduction

NLRP3 is one of the highly conserved pattern recognition receptors that plays role in recognition of intracellular pathogens and endogenous byproducts of tissue injury. Diverse stimuli, including nigericin, uric acid crystals, amyloid- β fibrils and extracellular ATP can activate NLRP3 inflammasomes. Upon stimulation, cytosolic NLRP3 subunits form an oligomerized inflammasome complex. Inflammasomes activate inflammatory caspases such as caspase-1, which processes proinflammatory cytokines interleukin 1 β (IL-1 β) and IL-18 for their maturation and cleaves gasdermin D to generate a N-terminal fragment to induce pore formation, cytokine release and pyroptotic cell death. Pro-inflammatory cytokines IL-1 β and IL-18 are associated with various malignancies.

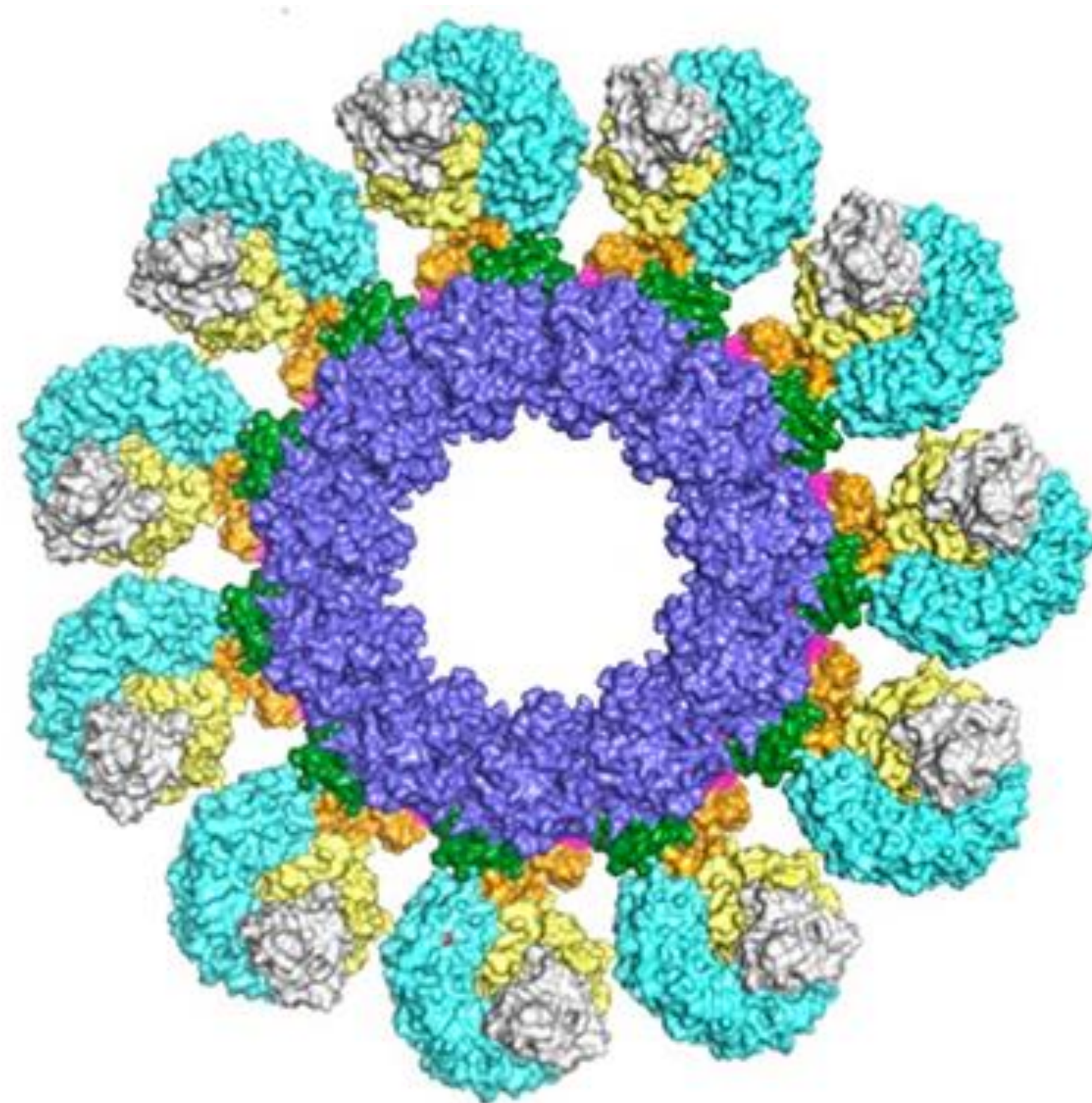


Figure 1: NLRP3 inflammasome complex

In the study, activation of the inflammasome with priming and activation signals will identify exclusive interacting proteins of cellular pathways. Identified genes will be analyzed to reveal their functions based on biological processes, molecular functions and cellular components. Phenotypic properties of monocytic cells after the treatment with LPS and MSU and or Nigericin will be observed.

Materials and Methods

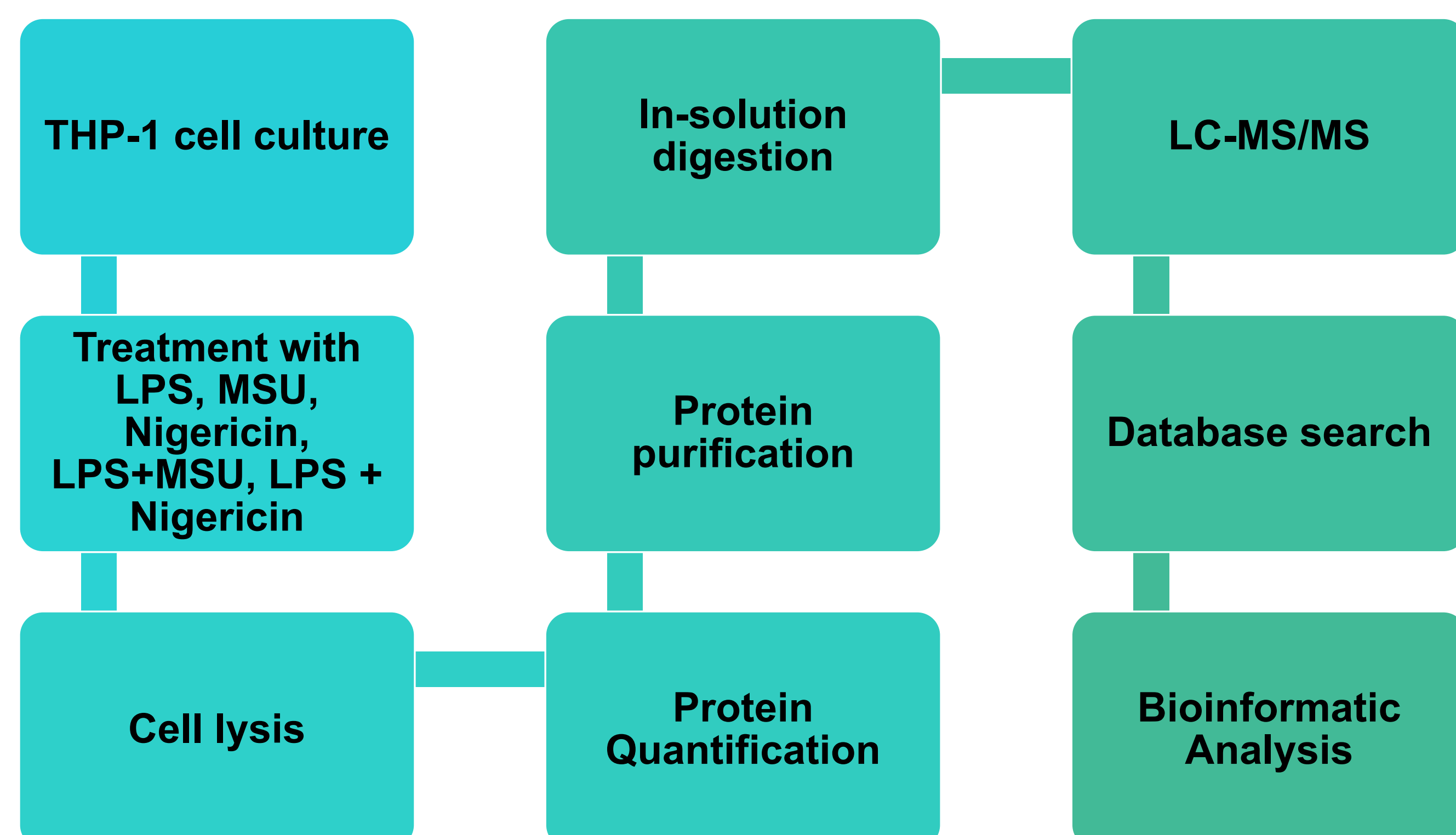


Figure 2: Proteomic profiling of THP-1 monocyte cells upon activation of inflammasome

Results

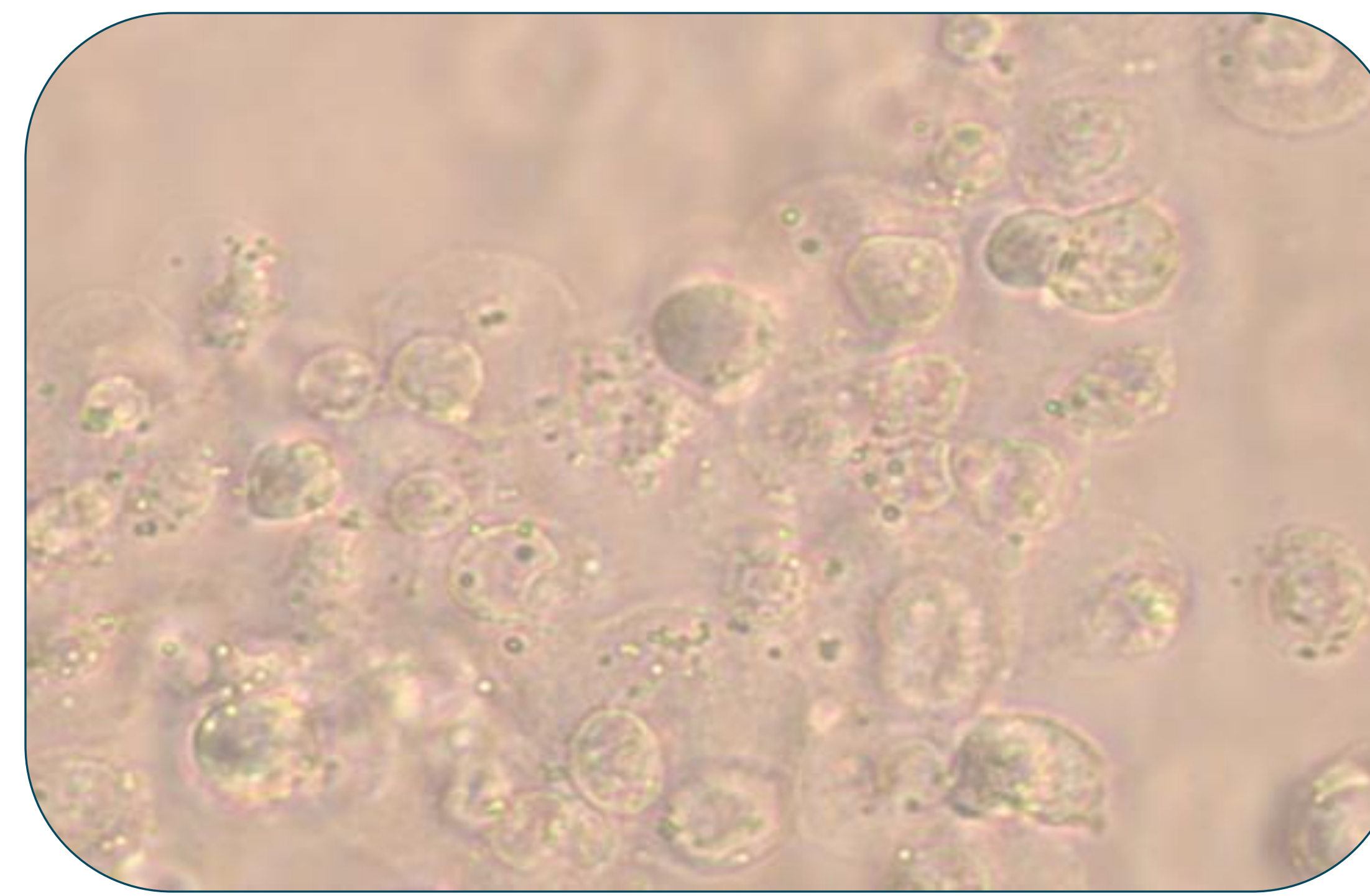


Figure 3: THP-1 cells upon treatment with LPS and Nigericin

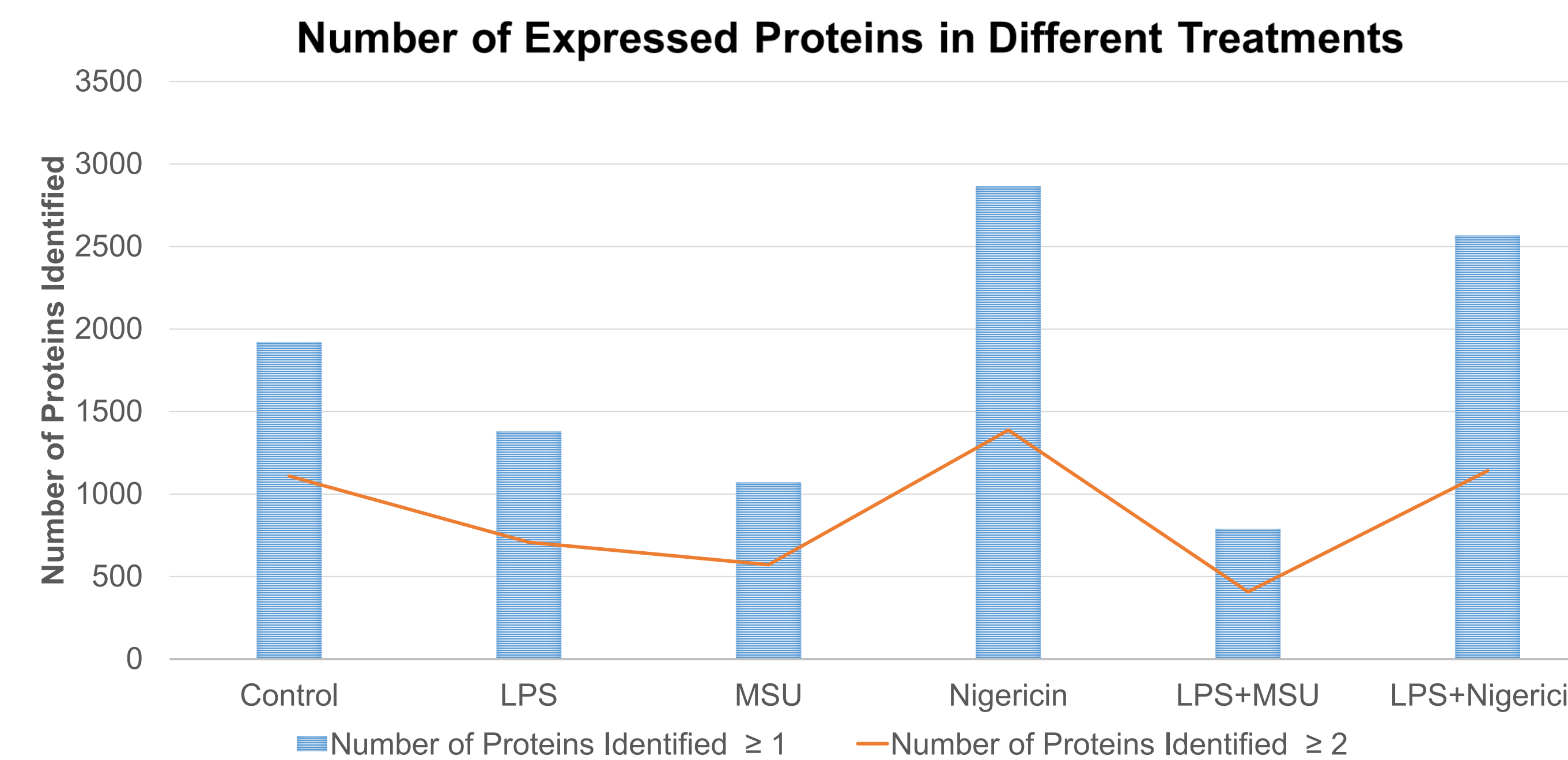


Figure 4: Identified proteins in different treatments

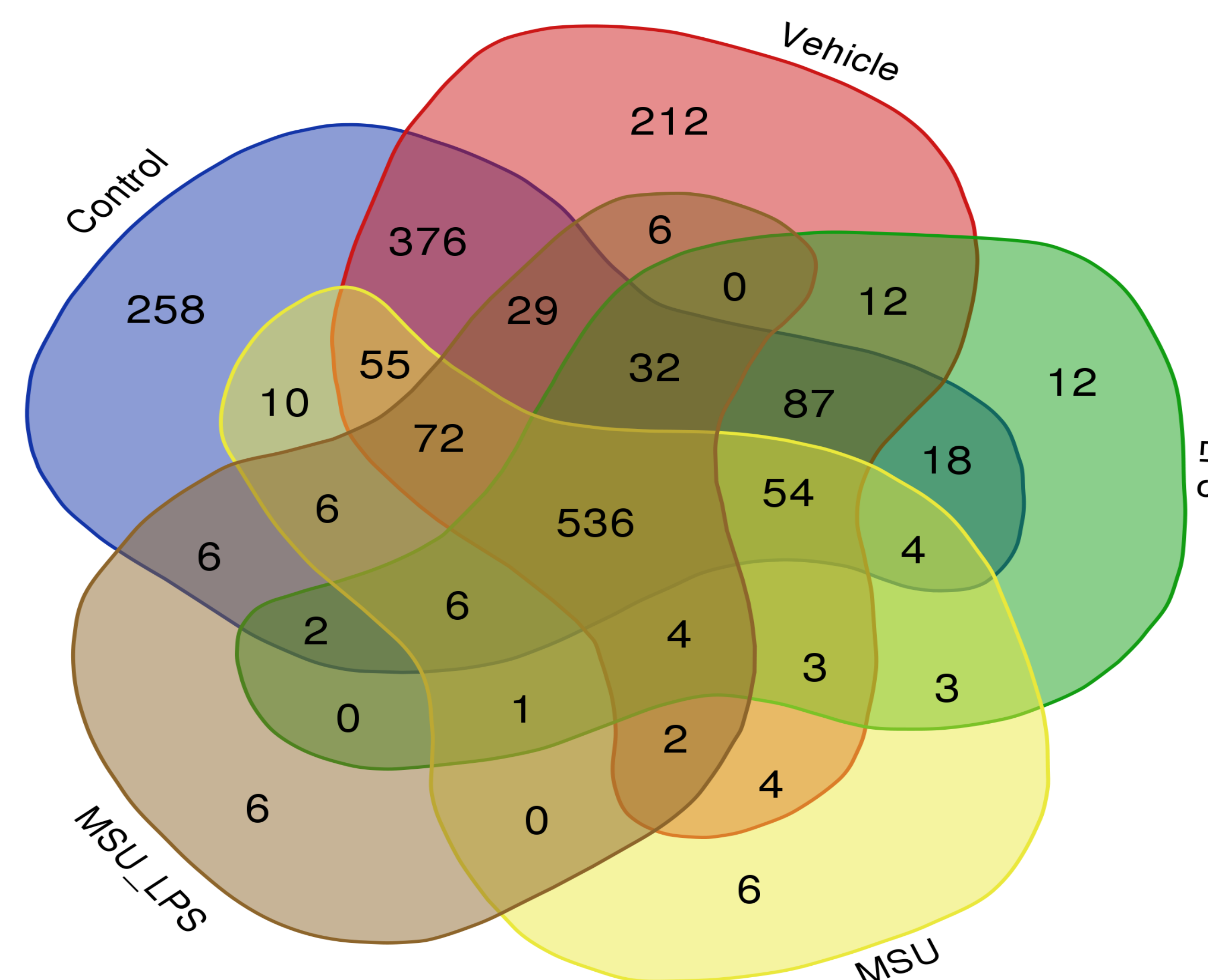


Figure 5: Venn diagrams of identified proteins in different treatments

Results

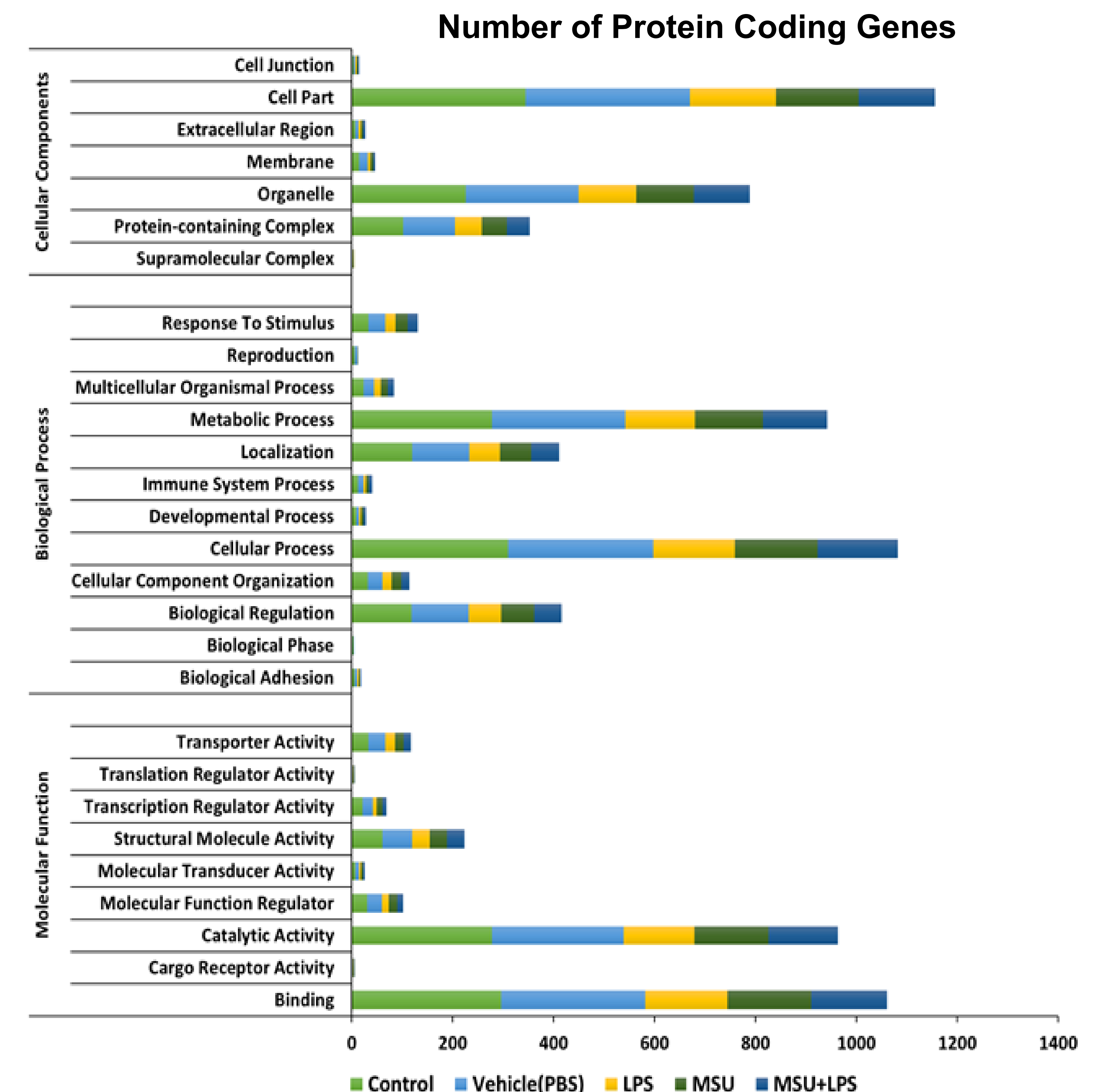


Figure 6: Analysis of gene ontology based on biological processes, molecular functions, and cellular components using the PANTHER classification system

Conclusion

- The proteomics approach identified six proteins that are uniquely expressed in NLRP3 activated macrophage cells.
- Proteins of the interactome mainly participate in catalytic and binding processes.

Future Direction

- Novel communicators of NLRP3 interactome can be identified with the use of affinity purification crosslinking mass spectrometry (AP-CXL-MS).

Reference

- Guo H, Callaway JB, Ting JP. Inflammasomes: mechanism of action, role in disease, and therapeutics. *Nat Med.* 2015;21(7):677-687. doi:10.1038/nm.3893
- Sharif H, Wang L, Wang WL, et al. Structural mechanism for NEK7-licensed activation of NLRP3 inflammasome. *Nature.* 2019;570(7761):338-343. doi:10.1038/s41586-019-1295-z

Acknowledgement

