



Serum-based untargeted metabolomics to investigate possible indicators of asymptomatic Lymphatic Filariasis

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INTRODUCTION

- Despite ongoing efforts to eradicate it, Lymphatic Filariasis (LF) remains a public health concern.
- Approximately 657 million individuals across 39 nations are still at risk, with India representing 62% of the worldwide at-risk population.
- LF must be diagnosed early to prevent permanent lymphatic damage. Despite their usefulness, current diagnostic methods have a number of drawbacks such as lack of specificity or sensitivity at early infection stages.
- Identifying biomarkers specific to the microfilaremic stage of the disease would facilitate early diagnosis and prevent progression to the more debilitating chronic stages.
- Serum based metabolomics is the extensive high throughput examination of small-molecule metabolites present in blood serum.

AIM: This study aims to detect host-derived serum metabolic and biochemical markers linked to the asymptomatic microfilaremic stages of LF by high resolution accurate mass spectrometry (HRAMS)

METHODOLOGY

Blood sample collection from Microfilaremic (84) and Normal (48) subjects from endemic locations of Varanasi (Field-based) and Filaria Centre

Blood serum separation and HRAMS analysis

- Blood is allowed to Clot
- Serum pipetted out
- Centrifugation to get clear serum devoid of cells
- Serum transferred to pre-labelled eppendorf tubes, stored at -20°C
- Global metabolomics (UP-LC-HRAMS)
- Data Analysis and Interpretation (with Compound Discoverer 3.3.3.20 and MetaboAnalyst 6.0)
- Normal and microfilaremic groups were compared using a two-tailed Student's t-test. The false discovery rate (FDR) Benjamini–Hochberg correction adjusted the p-value. Statistical significance was defined as p-value <0.05 .



Figure 1: A picture showing field sampling activities for blood collection in the Munari village of Varanasi

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REFERENCES

- Jabir, M., Thiruvengadam, K., Prabhakaran, P. S., Samuel, M. S., & Rahi, M. (2026). Exploring the landscape of lymphatic filariasis research in India: A scientometric analysis of two decades from 2000 to 2024. *PLOS Neglected Tropical Diseases*, 20(1), e0013908.
- Kumar, V., Mishra, A., Yadav, A. K., Rathaur, S., & Singh, A. (2022). Lymphatic filarial serum proteome profiling for identification and characterization of diagnostic biomarkers. *Plos One*, 17(7), e0270635.
- Dierks, C., Tober-Lau, P., Veletzky, L., Wang, Z., Zühlke, B., Ludwig, D., ... & Kurth, F. (2025). Plasma Proteomics Reveals Distinct Signatures in Occult and Microfilaremic Loa loa Infections. *The Journal of Infectious Diseases*, 232(3), e383–e392.
- Chienwichai, P., Thiangtrongjit, T., Tipthara, P., Tarning, J., Adisakwattana, P., & Reamtong, O. (2023). Untargeted serum metabolomics analysis of Trichinella spiralis-infected mouse. *PLOS Neglected Tropical Diseases*, 17(2), e0011119.

RESULTS & DISCUSSION

161
DEMs

24 up
regulated

137 down
regulated

Arachidonic acid,
Prostaglandins, Retinal,
etc

FIG 1: Volcano plot: Differential expression

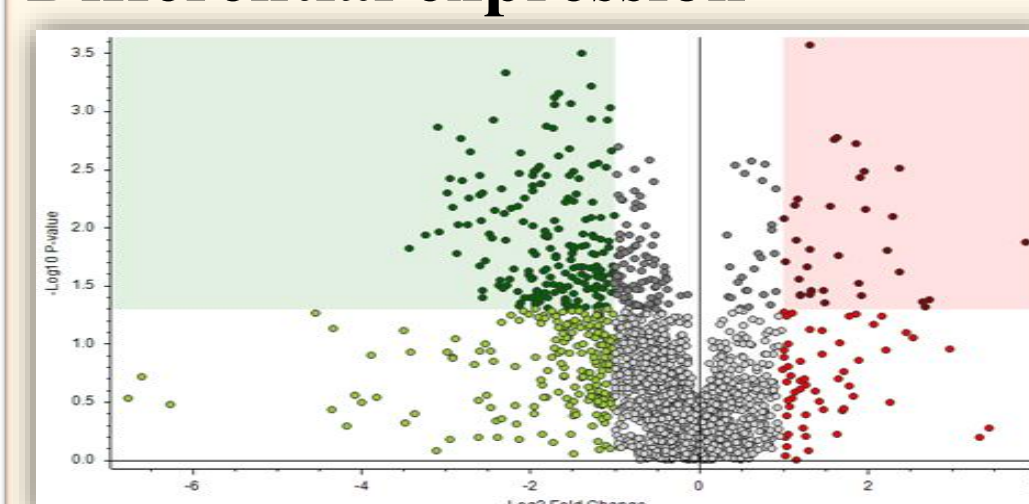
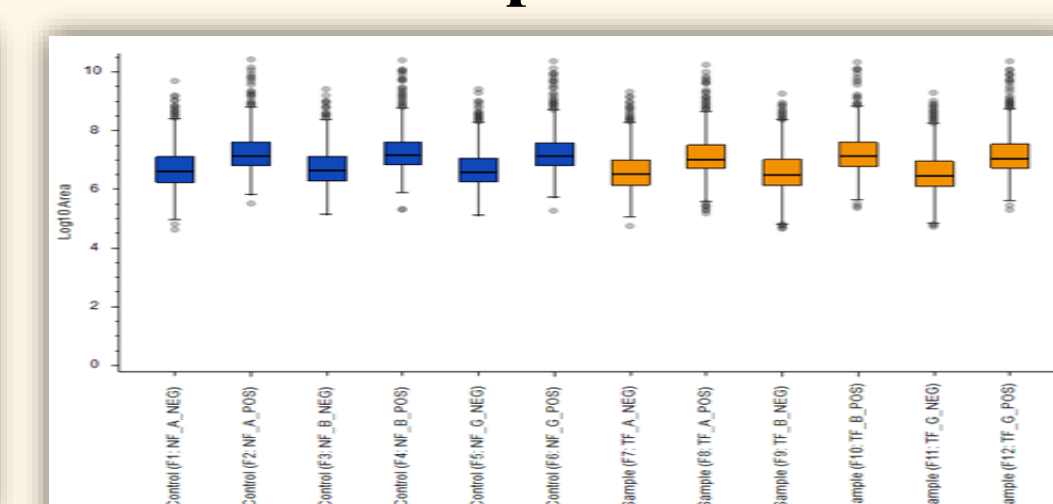


FIG 2: Box plot: Differential expression



Volcano plot showing downregulated and upregulated metabolites in MF groups ($p < 0.05$) in green and red, respectively.

FIG 3: PLS-DA and VIP score plot in Pos and Neg ESI modes

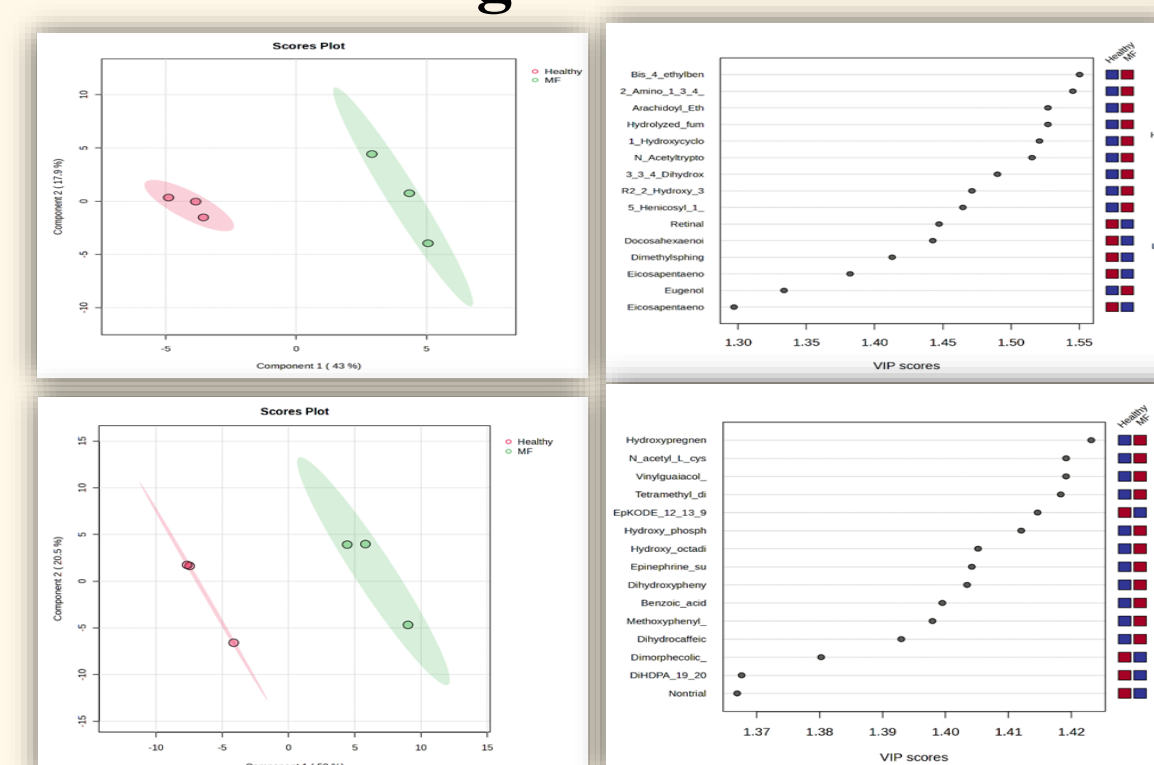
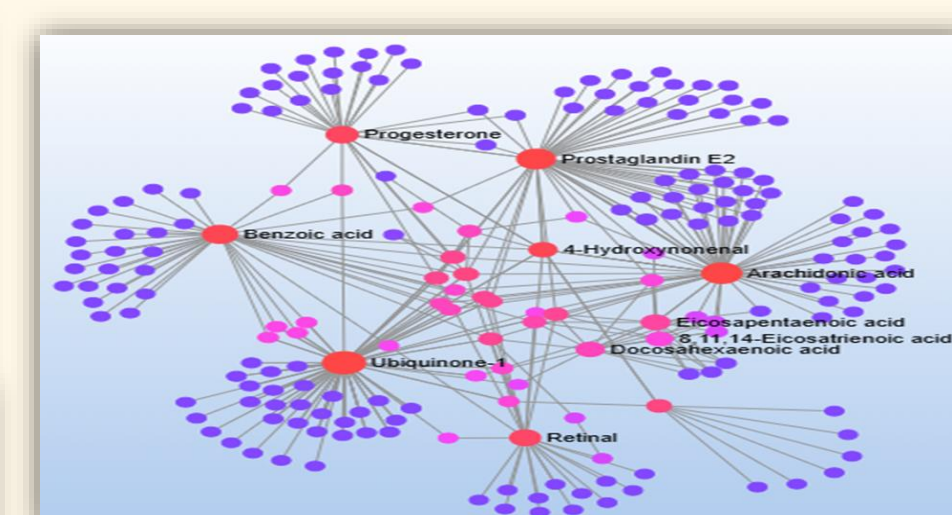


FIG 5: Metabolite-metabolite interaction network



Maximum interaction between metabolites shown in red bubbles indicating strong regulatory and/or functional relationships.

FIG 4: Pathway Analysis

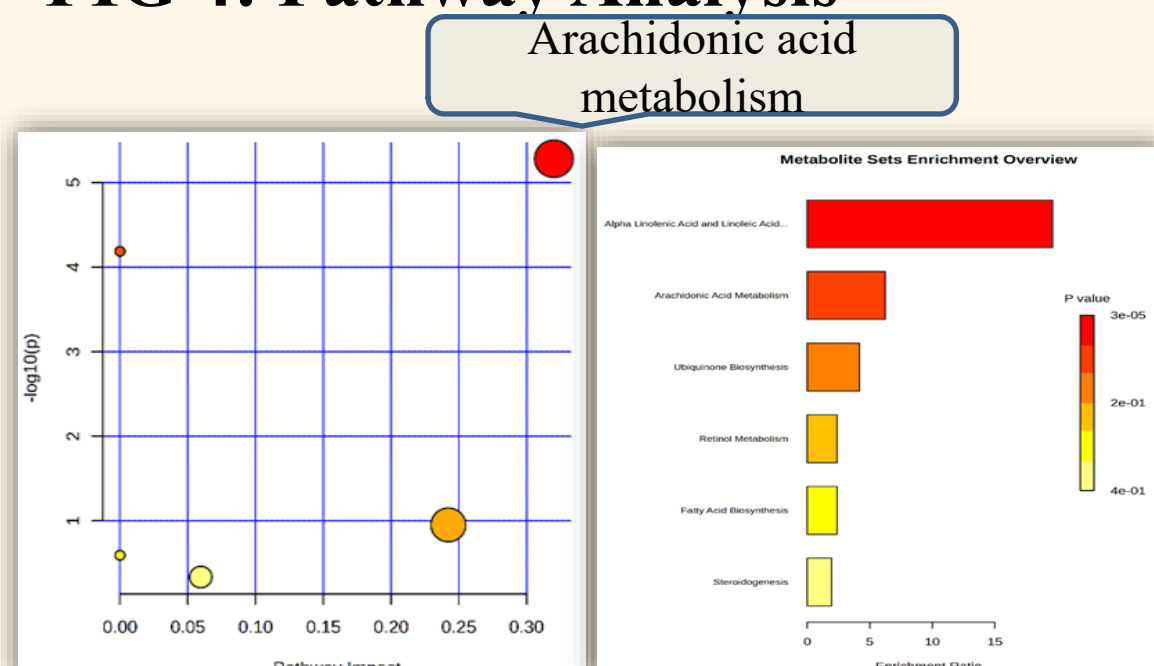
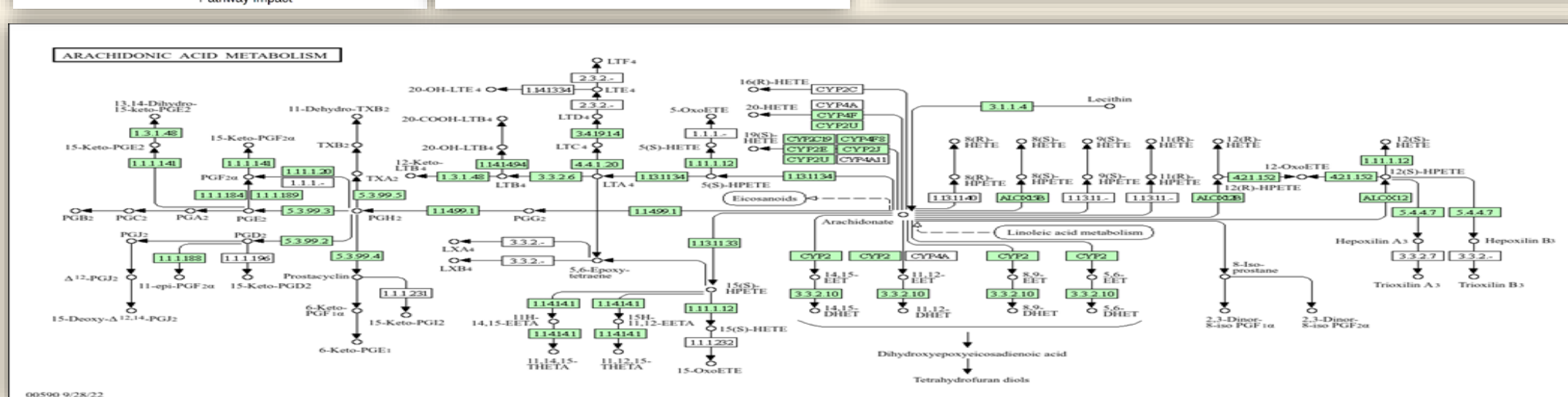
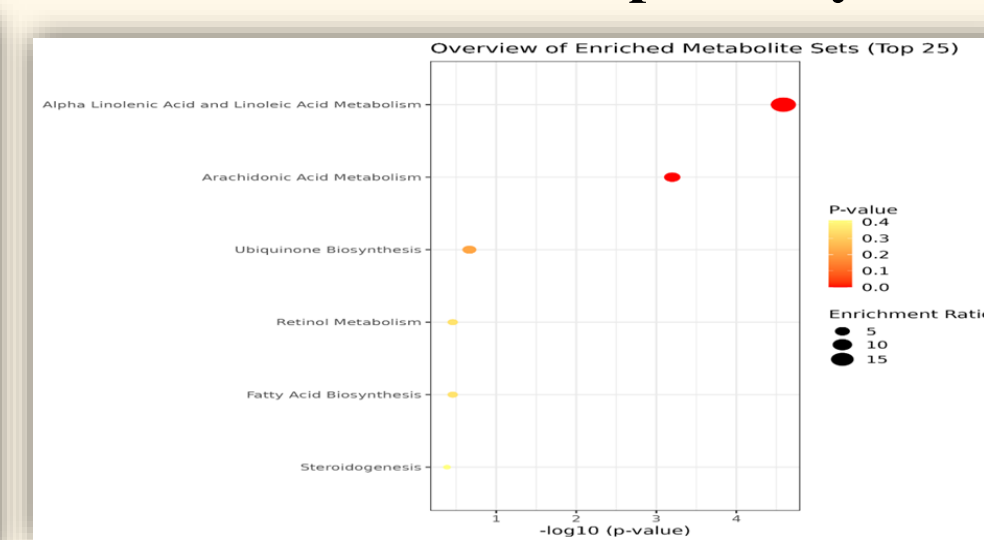


FIG: 6 Most significantly enriched metabolic pathways



KEGG'S metabolic pathway for Arachidonic acid

CONCLUSIONS

- HRAMS-based serum metabolomics identified 161 differentially expressed metabolites (DEMs), out of which 24 exhibited significant upregulation and 137 exhibited downregulation.
- Univariate and Multivariate discriminant analysis clearly distinguished microfilaremic individuals from healthy controls.
- Pathway analysis showed alterations in the metabolism of Arachidonic acid and unsaturated fatty acid synthesis.

FUTURE WORK

- Validation is required in larger independent cohorts.
- Future efforts will focus on targeted analyses to identify specific biomarkers for clinical translation.