

Benefiting From a Pest- *Paraupsylla Tuberculata* Infested *Alstonia Scholaris* Latex Cross-Reactivity To HCV, Plasmodium

Sriram Kannan¹, Deblina Mitra², Anierudhe Venkatavaradhan³,
Sreevani Kannan⁴

¹Adhoc Faculty Alumina, Rural Technology, Guru Ghasidas university, Bilaspur, Chhattisgarh (current affiliation ICMR)

²Teacher, Biology, TIS, Delhi

³Student Alumina, BS Abdur Rahman College, Chennai

⁴Associate Professor, Department of Physics, Panimalar Engineering College, Chennai

Keywords: *Paraupsylla tuberculata*; *Alstonia scholaris*; Plasmodium, HCV, Leaf Gall

Abstract

Alstonia scholaris is well known for its medicinal properties. Its bark is used for anti-malarial compounds and its leaf is used for treatment of dysentery. It is infested by the insect pathogen causing leaf gall namely *Paraupsylla tuberculata*. In this article we show for the first time that the latex of *Alstonia scholaris* has cross-reacting proteins with human pathogens namely HCV, Malaria and these cross-reacting proteins react differentially when infested by the insect pest *Paraupsylla tuberculata* causing leaf gall. Taking into consideration that plant galls are consumed by certain population, this finding could be important for underlying factor for resistance to certain diseases in certain group of people.

Introduction

Consumption of indigenous food and different variety of food is linked to combating chronic disease conditions (1) and also significant to combat food security issues (2). Consumption of at least 65 species of insects is known among the tribal people of Arunachal Pradesh (3). It is also known that plant with gall disease is used for medicinal properties (4,5,6). Folklore medicine is known to use plant galls (7). One such disease is the leaf gall disease in the wide spread tree found in India *Alstonia scholaris* also called as the devil's tree. It is known to be infested with *Paraupsylla tuberculata* that causes leaf galls which can be correlated to air quality index (8).

Supposedly consumption of leafgall renders immunity to disease as it is known that leaf galls are used for medicinal property, then characterising the proteins in that is the next step. One such report on zymogram of leaf gall of *Alstonia* has been reported (but the proteins have not been characterised (9). Taking into account, the rise in anti-microbial resistance with conventional antibiotics, novel drugs are being explored

(10,11,12), as well as novel candidate for vaccine. To study in depth any biological fluid in vivo, LASER methods have been described as for mouse blood, lesions (13).

Recently it has been shown that lateral flow assays coated with specific antigen or antibody of pathogens could be used as to screen the presence of cross-reactive plant proteins (14). Following that principle, the latex of *Alstonia* has been screened for HCV, Plasmodium with and without leaf gall disease as described in this article.

Materials and Methods

Lateral flow assay:

1. Rapid diagnostic kits were purchased for Plasmodium (Med source for Pf, Pv) and for HCV, HBV (Med source). The plant latex or sap were dropped at sample points of the lateral flow assay and the results were noted within 1-10' as soon as the control line turned positive. In case of HCV of Med source, the test line has recombinant antigens of HCV for detection of anti HCV antibodies.

2. Bioinformatic analysis: The phytochemical compound identified through IMPPAT database (<https://cb.imsc.res.in/impptat>) for a particular medicinal plant.

Results

The latex tested from leaves of *Alstonia scholaris* with leaf gall showed reactivity to HCV but latex from healthy leaves did not show (Figure 1). Contradictingly, the latex from leaves of *Alstonia scholaris* showed reactivity with *Plasmodium* (*Pf*, *Pv*) but the leaves with galls did not (Figure 1).

Discussion

Alstonia scholaris is known to have antimalarial compounds like Echitamine, tubotaiwine, akuamicine, picrinine, echitamidine, strictamine (15, 16) also indexed in the Indian medicinal plants, phytochemicals and therapeutics. Another compound detected through the IMPPT database is sinapic acid and this compound has shown to be antiviral as for SARS virus (17). As seen in figure 1, leaves that were uninfested displayed cross-reactivity to Plasmodium and it was absent in the infested leaf. The novelty is to screen the potential cross reacting protein present in the plant latex with the lateral flow assay. This indicates that the insect infestation alters the mimicry present either it introduces or eliminates mimicry. In case of HCV lateral flow, the plant latex indicated there was no cross-reactivity but after infestation with the *Paraupsylla tuberculata* resulted in cross-reactivity indicating the insect protein could have cross-reactivity or any new pathway in the leaf gall absent in a healthy leaf could have resulted in cross-reactivity with HCV.

Conclusion

Leaf extracts and Leaf galls are used for medicinal purposes but the molecular basis of it is unexplored. This article shows an example of cross-reactivity of leaf gall latex to human pathogen HCV and the leaf to Plasmodium.

References

1. Sarkar D, Walker-Swaney J, Shetty K. Food Diversity and Indigenous Food Systems to Combat Diet-Linked Chronic Diseases. *Curr Dev Nutr*. 2019 Sep 2;4(Suppl 1):3-11. doi: 10.1093/cdn/nzz099. PMID: 32258994; PMCID: PMC7101483.
2. Van Huis, A., Van Itterbeeck, J., Klunder, H., Mertens, E., Halloran, A., Muir, G., & Vantomme, P. (2013). *Edible insects: future prospects for food and feed security* (No. 171). Food and agriculture organization of the United Nations.
3. Chakravorty, J., Ghosh, S., & Meyer-Rochow, V. B. (2013). Comparative survey of entomophagy and entomotherapeutic practices in six tribes of Eastern Arunachal Pradesh (India). *Journal of Ethnobiology and Ethnomedicine*, 9, 1-12.
4. Eshwarappa, R. S. B., Ramachandra, Y. L., Subaramaihha, S. R., Subbaiah, S. G. P., Austin, R. S., & Dhananjaya, B. L. (2016). Anti-Lipoxygenase activity of leaf gall extracts of *Terminalia chebula* (Gaertn.) Retz.(Combretaceae). *Pharmacognosy Research*, 8(1), 78.
5. Eshwarappa, R. S. B., Iyer, R. S., Subbaramaiah, S. R., Richard, S. A., & Dhananjaya, B. L. (2014). Antioxidant activity of *Syzygium cumini* leaf gall extracts. *BioImpacts: BI*, 4(2), 101.
6. Birur Eshwarappa, R. S., Ramachandra, Y. L., Subaramaihha, S. R., Pasura Subbaiah, S. G., Austin, R. S., & Dhananjaya, B. L. (2016). Anti-Lipoxygenase Activity of Leaf Gall Extracts of *Terminalia chebula* (Gaertn.) Retz.(Combretaceae). *Pharmacognosy Research*, 8(1).
7. Patel, S., Rauf, A., & Khan, H. (2018). The relevance of folkloric usage of plant galls as medicines: Finding the scientific rationale. *Biomedicine & Pharmacotherapy*, 97, 240-247.
8. Talukdar, P., Das, K., Dhar, S., Talapatra, S. N., & Swarnakar, S. (2016). Galls on *Alstonia scholaris* leaves as air pollution indicator. *World Scientific News*, (52), 181-194.
9. Saini, D., & Sarin, R. (2012). SDS-PAGE analysis of leaf galls of *Alstonia scholaris* (L.) R. *Br. J Plant Pathol Microbiol*, 3, 121.
10. Maruthapandi, M., Das, P., Saravanan, A., Natan, M., Banin, E., Kannan, S., ... & Gedanken, A. (2021). Biocompatible N-doped carbon dots for the eradication of methicillin-resistant *S. aureus* (MRSA) and sensitive analysis for europium (III). *Nano-Structures & Nano-Objects*, 26, 100724.
11. Itshak-Levy, D., Israel, L. L., Schmerling, B., Kannan, S., Sade, H., Michaeli, S., & Lellouche, J. P. (2020). Disaggregation, stabilization, and innovative functionalization/surface engineering of detonation nanodiamonds via ultrasonication-promoted ceric ammonium nitrate treatment. *Diamond and Related Materials*, 104, 107738.
12. Kannan S, Harel Y, Levy E, Dolitzky A, Sagiv AE, Aryal S, Suleman L, Lellouche JP, Michaeli S. Nano-Leish-IL: A novel iron oxide-based nanocomposite drug platform for effective treatment of cutaneous leishmaniasis. *J Control Release*. 2021 Jul 10;335:203-215. doi: 10.1016/j.jconrel.2021.05.019. Epub 2021 May 19. PMID: 34019947.
13. Yariv I, Kannan S, Harel Y, Levy E, Duadi H, Lellouche JP, Michaeli S, Fixler D. Iterative optical technique for detecting anti-leishmania nanoparticles in mouse lesions. *Biomed Opt Express*. 2021 Jun 28;12(7):4496-4509. doi: 10.1364/BOE.425798. PMID: 34457428; PMCID: PMC8367277.
14. Kannan, S., Mitra, D., Kannan, S., & Venkatavaradhan, A. Lateral Flow Immunoassay As Novel Screening Tool For Potential Cross-Reacting Protein In Crude Plant Latex As Tested For Plasmodium, Dengue, HBV, HCV, HIV. *IJFMR-International Journal For Multidisciplinary Research*, 5(4).

15. Pankti, K., Payal, C. M. G. & Jagadish, K. (2012). A phytopharmacological review of *Alstonia scholaris*: A panoramic herbal medicine. *International Journal of Research in Ayurveda and Pharmacy*, 3(3), 367-371.
16. Ihwan & Koda, S. H. A. (2017). Antimalarial Herbal Plants in Kupang, Indonesia. *Biosaintifika: Journal of Biology & Biology Education*, 9(1), 95-104.
17. Orfali R, Rateb ME, Hassan HM, Alonazi M, Gomaa MR, Mahrous N, GabAllah M, Kandeil A, Perveen S, Abdelmohsen UR, Sayed AM. Sinapic Acid Suppresses SARS CoV-2 Replication by Targeting Its Envelope Protein. *Antibiotics (Basel)*. 2021 Apr 11;10(4):420. doi: 10.3390/antibiotics10040420. PMID: 33920366; PMCID: PMC8069661.

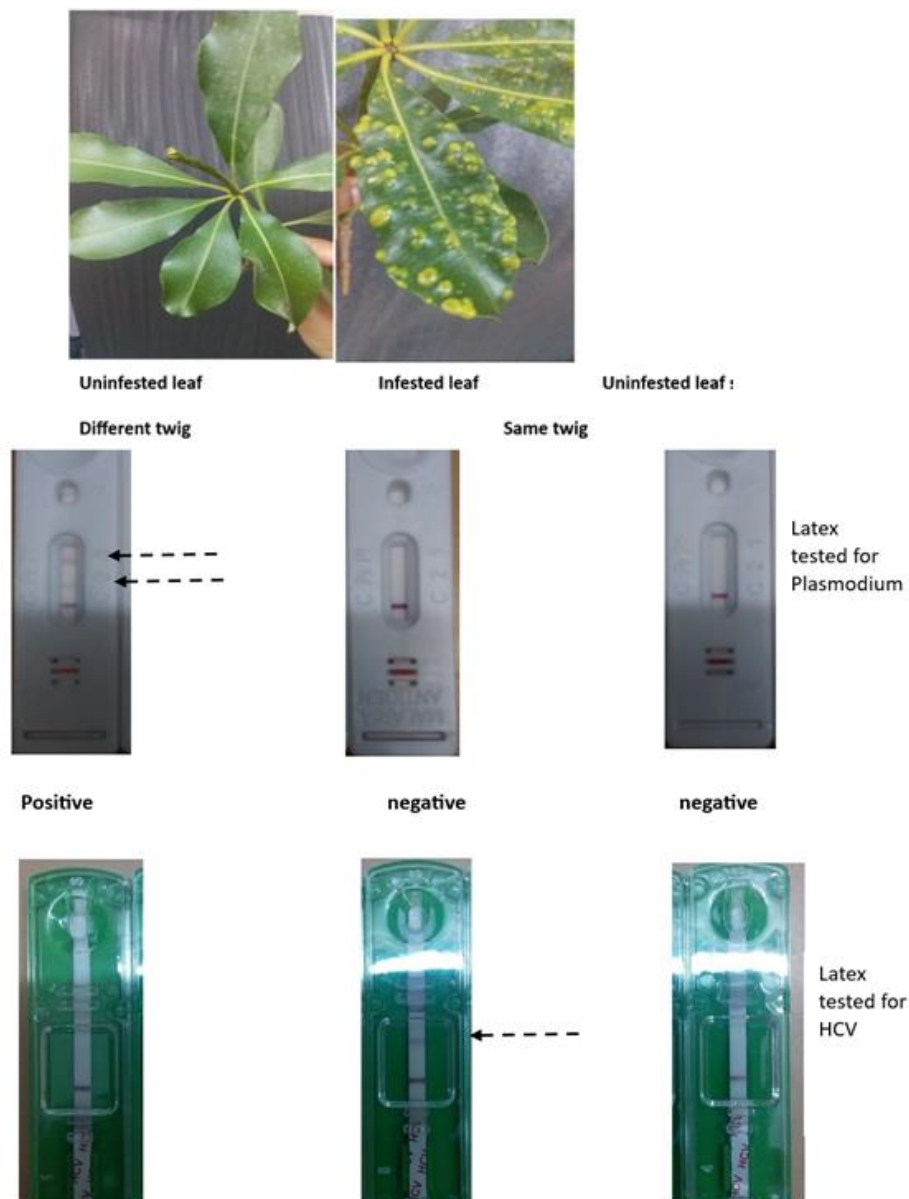


Figure 1 Lateral flow assay on infested and uninfested leaves

Representative image of lateral flow assays (Med source) with the latex of infested and uninfested latex from leaf of *Alstonia*