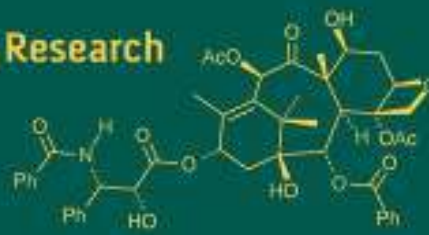
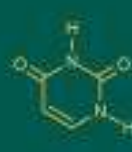


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GC-MS based metabolomic profiling of sandalwood induced systemic signalling in host plants through a split root system

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Abstract

Indian sandalwood (*Santalum album* L.) is a xylem-tapping hemiparasitic tree that relies on host plants for water and mineral nutrients. Although post-haustorial nutrient transfer has been well documented, the initial signaling associated with host recognition and systemic responses are still not well understood. The current study used the hydroponic split root system to spatially segregate the root systems of three host species (*Mulberry*, *Casuarina*, and *Alternanthera*) and tested whether exposure to sandalwood would induce systemic signalling in physically separated root systems. Under control conditions, where the roots of the hosts were evenly distributed in two compartments without sandalwood, the metabolic profiles demonstrated the stable and homogeneous pattern of exudation. 37 metabolites were found to be common in the exudates of the roots of *Mulberry*, while 39 metabolites were found to be common in the exudates of *Casuarina* and *Alternanthera*. However, when sandalwood was placed in the left compartments, significant reprogramming of the metabolic profile was observed. When the roots of *Mulberry* were placed in the presence of sandalwood, the number of common metabolites was found to have reduced drastically to only 5, while unique metabolites were found in the non-exposed and exposed roots. *Casuarina* showed reduced numbers of common metabolites to 9, while 15 unique metabolites were found in the non-exposed roots. *Alternanthera* showed similar responses. This indicates the systemic signalling from exposed roots to non-exposed roots system and strong metabolic reprogramming. KEGG annotation supported the presence of L-lysine (C00047) in the root exudates of *Casuarina*, which was involved in degradation (map00310), amino acid biosynthesis (map01230), and secondary metabolisms, including alkaloid biosynthesis (map01064). The present study shows that sandalwood induces a rapid, long-distance metabolic shift in the host root systems regardless of physical exposure, suggesting the perception of chemical sandalwood cues and the activation of root to root signalling.

Keywords: *Santalum album*, Split root system, GCMS, KEGG pathway

Introduction

Indian sandalwood (*Santalum album* L.) is a central tropical hemiparasitic arboreal organism, which is very widespread in tropical and subtropical regions. Although the plant has photosynthetic capacities of its own, its growth pattern and developmental stages have been found to be partly dependent on the heterotrophic relationships with host flora, whereby it acquires water and mineral nutrients needed. The genus *Santalum* is placed taxonomically among the cadres of xylem tapping hemiparasites, with the expression of specialized invasive organelles located as haustoria (Mallaburn, 1992) [20]. The structures actively inch through host root tissues and establish vascular continuity that allows the free flow of resources between parasites and hosts (Tennakoon *et al.*, 1997) [33]. In *S. album*, haustorial connections may also be established with nitrogen-fixing nodules in the case of leguminous hosts, thus improving nutrient uptake (Lu *et al.*, 2014) [19].

Sandalwood has the ability to associate with a wide range of host species, however, it has been shown to have a particular association with some of the chosen hosts, which include *Acacia*, *Casuarina*, *Dalbergia* and *Sesbania*. Previous studies by Lu *et al.* (2014) [19] has determined that the relationship with nitrogen fixing hosts significantly elevates the amount of nitrogen in leaves, photosynthetic performance, and water-use performance in sandalwood. Besides, stable-isotope experiments have been used to support a two-way exchange of carbon and nitrogen between *S. album* and nitrogen-fixing hosts, thus showing

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complex physiological integration (Lu *et al.*, 2014) ^[19]. Lastly, it has been shown that the strength of the host-parasite interaction varies with nitrogen availability, suggesting a central role of the nutritional condition of the host in regulating the strength of this interaction (Meng *et al.*, 2021) ^[24]. The study by Anil and co-workers has shown that non-leguminous *Casuarina* is a superior host and induces enhanced sandalwood growth in plantations (Mohapatra and Anil, 2022) ^[26].

The early phases of host recognition and successful infection during the complex life cycles of parasitic angiosperms are largely mediated by chemotactic signals released by the roots of the host species (Kubo *et al.*, 2009) ^[18]. Notably, the empirical evidence suggests that about 70% of the sandalwood seedlings can begin haustorial formation after about 30 days of germination, reflecting the extraordinarily early onset of host dependent morphogenesis (Nagaveni and Srimathi, 1985) ^[27]. Verma *et al.* (2024) ^[36] revealed that physiological processes and antioxidant systems in sandalwood were mainly governed by the host species. Even though the pathways of nutrient translocation during post attachment are relatively well understood, the complex processes that govern pre-attachment host detection and the early signalling interactions remain poorly characterized.

The split root *in vitro* system (SRS), that physically partitions the root of a plant into two compartments, offers a complex methodological approach to the breakdown of local and systemic physiological activities in an individual plant root system. The method has been massively used to understand plant signalling networks (Torres *et al.*, 2021) ^[34], sectoriality, which is the compartmentalization of resource and hormonal fluxes within discrete physiological units in a plant (Marquis, 1996; Orians *et al.*, 2004) ^[23, 28] and water uptake (Ameglio *et al.*, 1999), ion transport (Gloser *et al.*, 2009b) and ectomycorrhizal interactions (Rose *et al.*, 2023) ^[11]. In addition to the physiological research, the split-root system (SRS) method has been used in ecological research to investigate plant - plant interactions and plant - microbe interactions. The SRS method has been used to investigate the dynamics of interspecific competition (Marler, 2013) ^[22] as well as plant interactions with pathogenic organisms and mycorrhizal associations

(Kennedy *et al.*, 2009) ^[17]. The flexibility of the SRS allows for the distinction between contact and systemic signaling responses in the same plant.

Although many studies have explained how nutrients move from the host to sandalwood after the haustorium was formed, it is still not understood whether chemical signals released by sandalwood can induce metabolic changes in the host plants before the formation of direct haustoria connection. This study aims to check whether the chemical signals perceived in one part of the root network of one host could be transmitted to spatially separate root compartments, and thereby, induce metabolic reconfiguration in the absence of direct physical contact with sandalwood root exudates

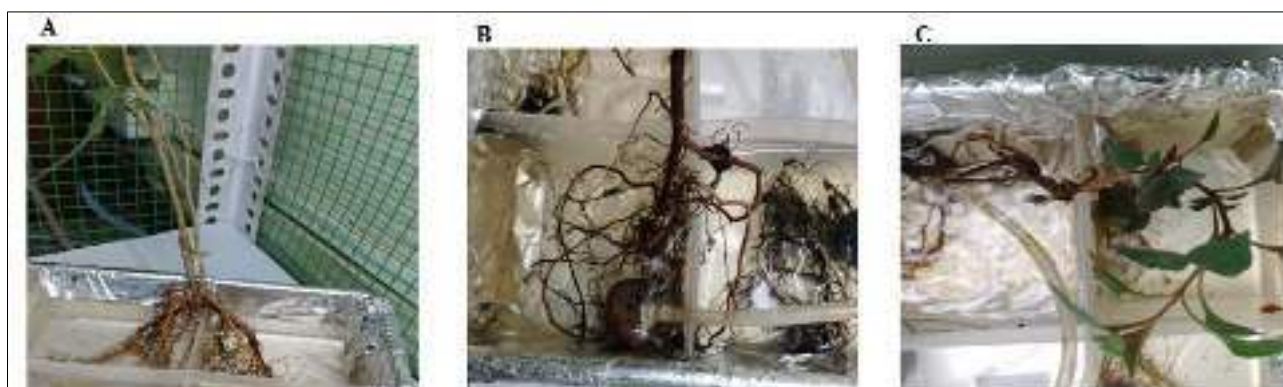
Materials and methods

The split root experiment was carried out at the Department of Plant Biotechnology, University of Agricultural Sciences (UAS), GKVK, Bengaluru, to assess the metabolite variation caused due to sandalwood-host root interactions. Sandalwood (*Santalum album*) plantlets and *Casuarina* seedlings were collected from Tapovana Sandalwood Nursery, Bengaluru, while *Mulberry* plants were purchased from the Department of Sericulture, GKVK. The experiment was set up in hydroponic systems using Hoagland's nutrient solution in a controlled growth chamber with temperature, light, and humidity controls. The plants were first grown in Hoagland's solution for a period of two months to allow them to establish properly and acclimatize to the hydroponic environment. After the acclimatization process, the root system of each host plant was carefully split into two equal parts and placed in two separate compartments labelled as left (L) and right (R) (Table 1, Fig 1). In treatment combinations, sandalwood seedlings were placed in the left compartment along with half of the host root system, while the right compartment had only host roots. Control treatments had host plants raised in split root systems without sandalwood. The root exudates were collected six days after treatment setup for GCMS analysis. Root exudates in the hydroponic system were collected and subjected to solvent extraction using ethyl acetate. The extracts were filtered and concentrated prior to analysis.

Table 1: Treatment details of Split root experiment with hosts plants

Treatment name	Description
M(L) control - M(R) control	<i>Mulberry</i> without sandalwood in the Left (L) compartment
C(L) control - C(R) control	<i>Casuarina</i> without sandalwood in the first compartment
A(L) control - A(R) control	<i>Alternanthera</i> without sandalwood in the first compartment
M+S(L) - M(R)	<i>Mulberry</i> with sandalwood in the left compartment
C+S(L) - C(R)	<i>Casuarina</i> with sandalwood in the left compartment
A+S(L) - A(R)	<i>Alternanthera</i> with sandalwood in the left compartment

Note: L: left compartment; R: Right compartment; M: *Mulberry*; C: *Casuarina*; A: *Alternanthera*; S: Sandalwood



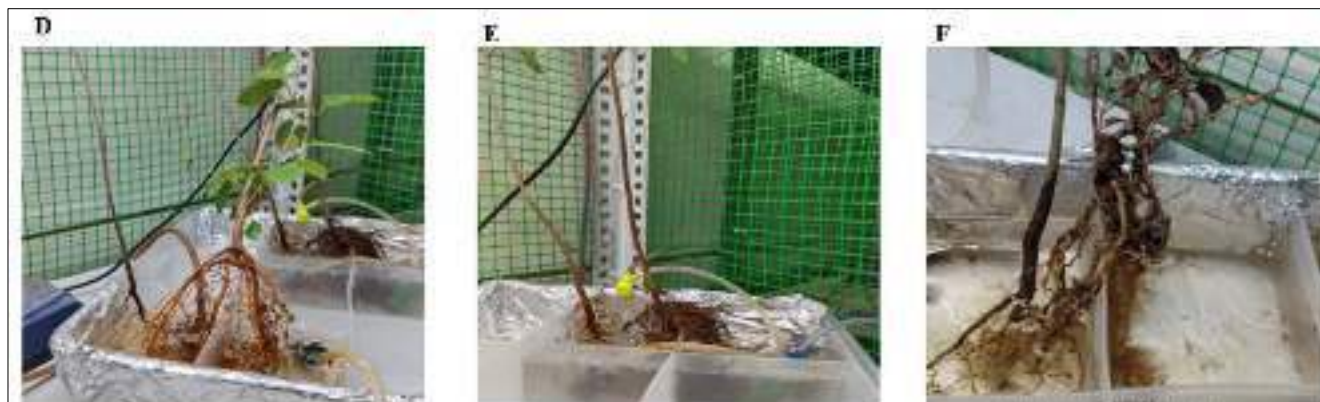


Fig 1: The split root set up for host plants grown with and without sandalwood plant. A) *Mulberry* without sandalwood, B) *Casuarina* without Sandalwood C) *Alternanthera* without sandalwood D) *Mulberry* with Sandalwood, E) *Casuarina* with Sandalwood, F) *Alternanthera* with Sandalwood

The prepared samples were analysed using a GC-MS system (QP2030, Shimadzu) to identify the bioactive compounds present in root extracts. Separation was achieved using an SH-RXI-5SIL MS capillary column (30m×0.25mm internal diameter, 0.25µm film thickness). Helium was used as the carrier gas at a constant flow rate of 1.0ml min⁻¹. The injector temperature was maintained at 280 °C. The oven temperature started at 50 °C and was held for 5 minutes. It was then gradually raised to 280°C, with intermediate hold points listed in Table 1. The entire temperature program cycle took 45 minutes to complete. The detailed oven temperature program used during GC-MS analysis is presented in Table 1. The mass spectrometer was operated in electron ionization (EI) mode at 70eV, with the ion source temperature maintained at 230 °C (Table 2). Compounds were identified based on their retention time and mass spectral data.

Table 2: Column temperature and oven program used in GC-MS analysis of root exudates

Rate (°C)	-	10.00	15.00	15.00	10.00	25.00
Temperature (°C)	50.0	100.0	150.0	200.0	220.0	280.0
Hold Time	5.00	5.00	5.00	5.00	3.00	5.00

Venn Diagram Analysis

To visualize the overlap and uniqueness of metabolites identified across different treatments, Venn diagrams were generated using the Venny 2.1 web tool. This tool enables rapid comparison of multiple datasets and facilitates identification of common and treatment specific compounds.

KEG pathway analysis

KEGG pathway analysis was carried out on metabolites identified by GC-MS. This helps us to understand the biological significance of the detected compounds. Analysis was carried out using MetaboAnalyst 6.0 to find the metabolic pathways that the target compounds in the root exudates affects. The compound's KEGG ID was mapped to the KEGG pathway database.

Results

GC-MS profiling of *Mulberry* and *Casuarina* root exudates in the split root experiment showed significant metabolic differentiation between control and sandalwood associated treatments.

In control treatment, where *Mulberry* roots were evenly divided in both compartments without sandalwood (M(L) control vs M(R) control), the metabolic profiles were very similar. A total of 37 metabolites were common in both compartments, while only 4 and 8 metabolites were unique to the left and right compartments, respectively (Fig 2B). In contrast, the introduction of sandalwood to the left compartment (M+S(L)) caused significant changes in the metabolite profile between the root exudates in left and right compartments. A three-way comparison among M+S(L), M(R), and the set of metabolites from the control treatment showed that only 5 metabolites were common to all. Interestingly, 8 metabolites were found exclusively in the right compartment (M(R)) when sandalwood was present in the left compartment (Table 3, Fig 2C). These included (S)-glyceraldehyde, a central intermediate in central carbon metabolism; growth-related hydrocarbons like 3,7-dimethyldecane; 1-heptadecene, a precursor for signalling molecule biosynthesis; antioxidant phenolics like 3,5-di-tert-butylphenol; the cuticular wax compound 2-methylhexacosane; and ethoxy (methyl) chlorosilane, involved in ethylene-related metabolic processes (Fig 2A). GC-MS analysis of *Casuarina* root exudates in the split-root experiment showed that, In the control treatment (C(L) control vs C(R) control), 39 metabolites were common to both, with only 9 unique metabolites found in each side (Fig 3B). This is a very high level of commonality and suggests a strong level of baseline metabolic homogeneity and exudation patterns in the absence of sandalwood interaction. After the addition of sandalwood to the left side (C+S(L)), the level of commonality in metabolite composition dropped significantly to 9 common compounds. Three-way comparison among C+S(L), C(R), and the control metabolite set showed that only 4 metabolites were common to all. Notably, 15 metabolites were found exclusively in the right compartment (C(R)) when sandalwood was restricted to the left compartment (Fig 3C). These included a hexahydropyridine derivative with known antioxidant activity; L-lysine, involved in protein synthesis and stress response; methoxyacetic acid, heptadecyl ester, with known herbicidal properties; hydrocarbons like 2-methyloctane and nonane derivatives, known to be involved in plant defense and cuticular wax composition; and 2-adamantanone oxime, a precursor of biologically active adamantane derivatives (Fig 3A).

Table 3: Compartment Specific Unique Compounds in Root Exudates of *Mulberry* in the Right compartment of a Split Root System Exposed to Sandalwood in the Left Compartment

Sl. No.	Compound	M/Z value	Biological Role(s) in Plants	Citation
1	Propanal, 2,3-dihydroxy-, (S)- (S)-Glyceraldehyde	90.08	Central intermediate in glycolysis, gluconeogenesis, and Calvin cycle	KEGG Compound: C00118, YMDB
2	Decane, 3,7-dimethyl-	170.34	Plant Growth Regulatory Activity	Cutler <i>et al.</i> , 1997 ^[8]
3	Phenol, 3,5-bis(1,1-dimethylethyl)-	206.32	Antioxidant	Miyazawa <i>et al.</i> , 2003 ^[25]
4	Ethoxy(methyl)chlorosilane	124.64		
5	2-Methylhexacosane	380.74	Component of cuticular wax; protection from desiccation	Jetter and Kunst, 2008 ^[14]
6	1-Heptadecene	238.45	Signalling molecule biosynthesis during seed germination and development.	Tao <i>et al.</i> , 2017 ^[32]
7	Histamine, N-BOC-5-iodo-	337.16		
8	Cetene (likely 1-tetradecene)	196.37		

Note: M/Z value: Mass to charge ratio

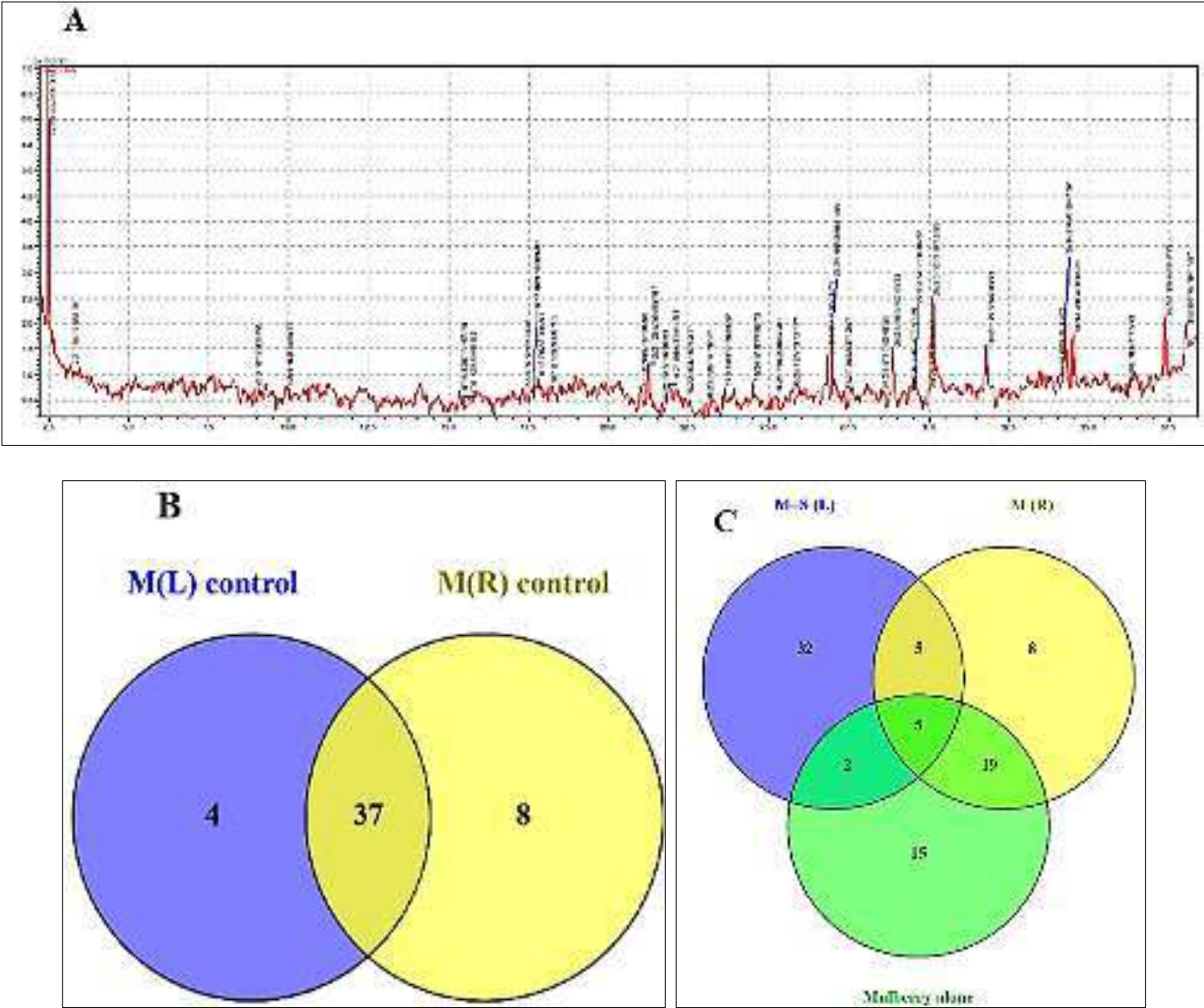
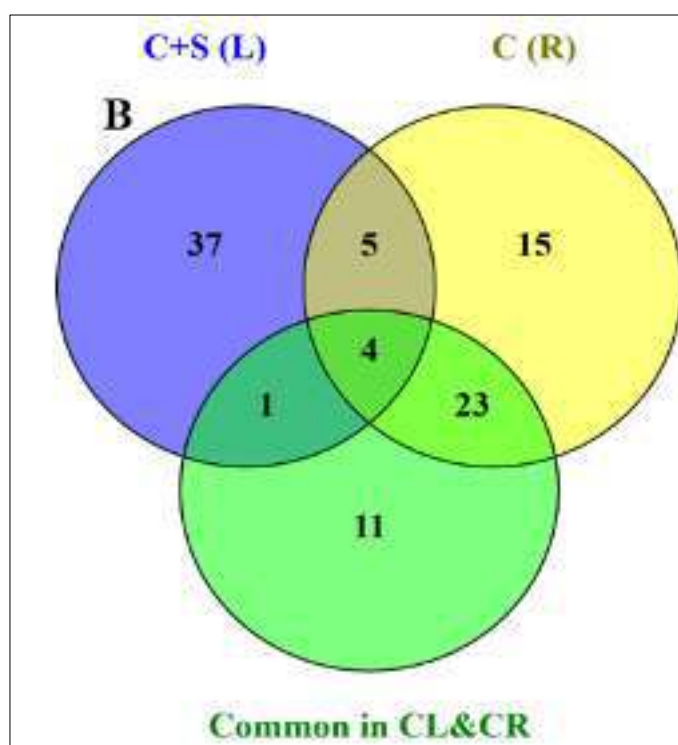
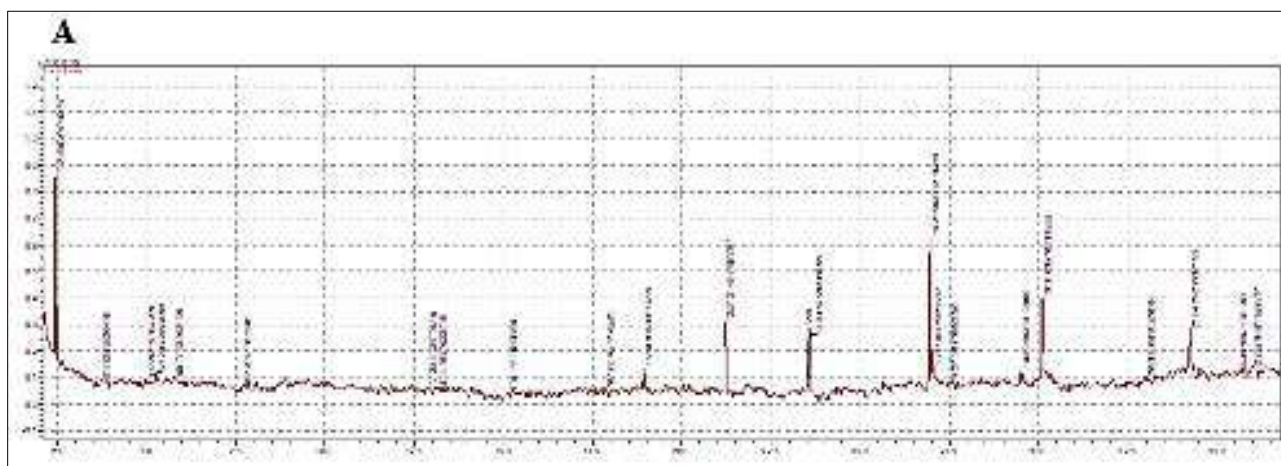


Fig 2: GCMS analysis of root exudates of *Mulberry* from split root experiment A) Chromatogram of GC-MS analysis of root exudates of M (R) compartment in split root system B) Venn diagram of comparing the metabolites from M(L) control and M(R) control C) Venn diagram of comparing the metabolites from M + S (L), M(R) and *Mulberry* alone

Table 4: Compartment Specific Unique Compounds in Root Exudates of *Casuarina* in the Right compartment of a Split Root System Exposed to Sandalwood in the Left Compartment

Sl. No.	Compound Name	Biological Role(s) in Plants	Citation / Source
1	2-Propanamine, N-ethyl-N-nitroso-		
2	Hexahydropyridine, 1-methyl-4-[4,5-dihydroxyphenyl]-	Antioxidant	Natural Product Reports, 2020
3	L-Lysine, 5-hydroxy-	Protein biosynthesis and stress Signalling	Veiga-da-Cunha <i>et al.</i> , 2012 ^[35]
4	Methoxyacetic acid, heptadecyl ester	Herbicidal Activity	Hughes <i>et al.</i> , 1971 ^[12]
5	Silane, trimethyl-[[[(17.alpha.)-19-norpregn-4-en-20-yn-17-yl]oxy]-		
6	4-Phenylazophenol, TBDMS derivative		
7	Nonane, 5-(1-methylpropyl)-	Component of waxes	Jetter and Kunst, 2008 ^[14]
8	Octane, 2-methyl-	Involved in plant defense	Dudareva <i>et al.</i> , 2013 ^[9]
9	Cyclohexasiloxane, dodecamethyl-		
10	Undecane, 4,8-dimethyl-		
11	Hexasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11-dodecamethyl-		
12	Hexadecane, 1-iodo-		
13	2-Adamantanone oxime	plant-plant interaction	Inderjit and Duke, 2003
14	3-Chloropropane-1,2-diol, bis(tert-butyldimethylsilyl)		
15	(1-Methyl-4-oxopyrazolo[3,4-d]pyrimidin-5-yl) acetate		

Note: M/Z value: Mass to charge ratio



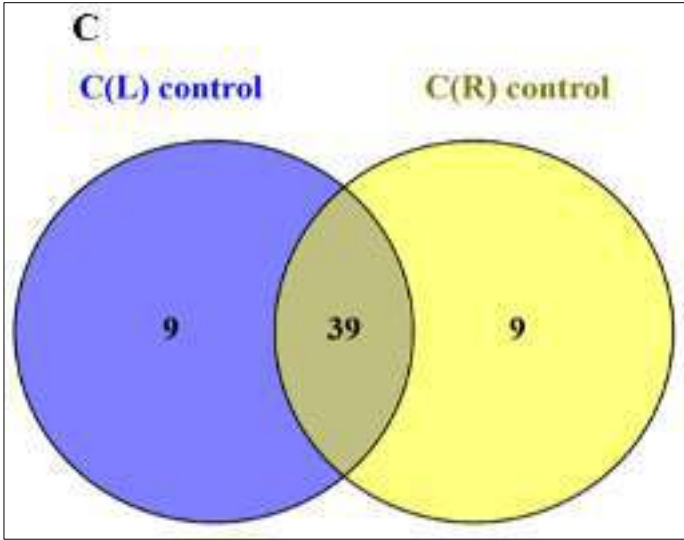
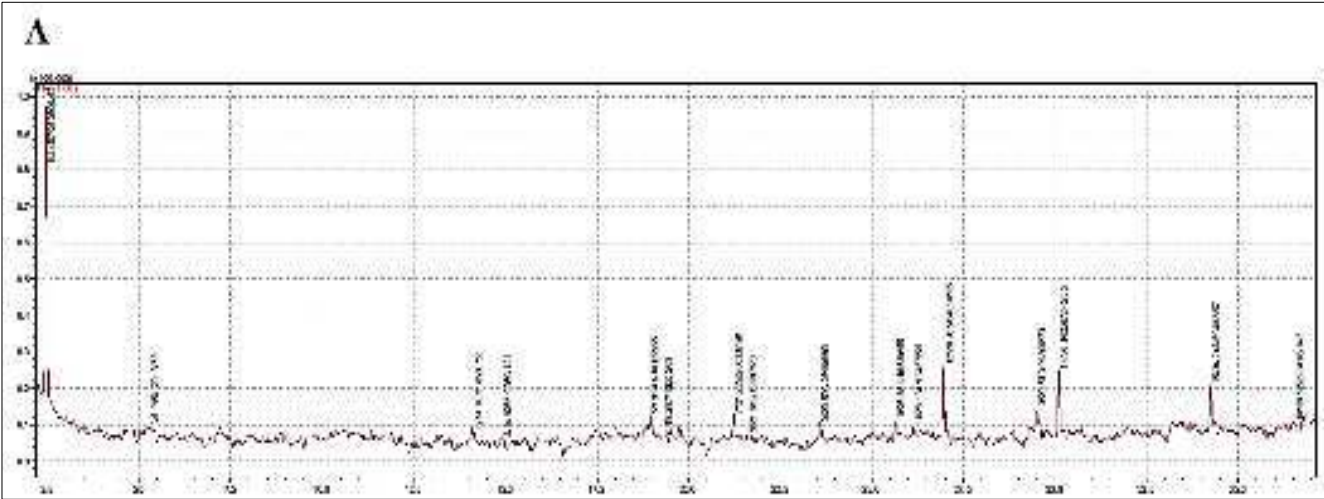


Fig 3: GCMS analysis of root exudates of *Casuarina* from split root experiment A) Chromatogram of GC-MS analysis of root exudates of C (R) compartment in split root system; B) Venn diagram of comparing the metabolites from C(L) control and C(R) control; C) Venn diagram of comparing the metabolites from C + S (L), C (R) and *Casuarina* alone

Table 5: Compartment Specific Unique Compounds in Root Exudates of *Alternanthera* in the Right compartment of a Split Root System Exposed to Sandalwood in the left Compartment

Sl. No.	Compound	Biological Role(s) in Plants	Citation
1	4H-Furo[3,2-b]pyrrole-5-carboxylic acid	Signalling or defense molecules in plant-microbe interactions	PubChem
2	3-(Trifluoromethyl)-[1,2,4]triazolo[3,4-b][1,3,4]thiadiazol-6-amine		
3	D-Arabino-Hexose, 2-deoxy-, cyclic 1,2-ethanediyl mercaptal, tetraacetate		
4	Propane, 3-chloro-1,1,1-trifluoro-		
5	2,4-Di-tert-butylphenol	Natural herbicidal compound,Induces lipid peroxidation in plants.	Aung <i>et al.</i> , (2020)
6	Hexane, 2,3,4-trimethyl-		
7	Eicosane	Component of plant cuticular waxes	Jetter and Schäffer, 2001
8	Uridine, 5-heptafluoropropyl-		
9	Acetic acid, chloro-, octadecyl ester		
10	Madumycin II	Antibiotic from Streptomyces; Act as endophyte metabolite	Managamuri <i>et al.</i> , (2020)
11	Thiourea, 1,1-diethyl-3-(3-methoxyphenyl)-		
12	2-Chloro-3-oxo-N-phenylbutanamide, TMS derivative		
13	Pentasiloxane, dodecamethyl-		
14	2-(Pyridin-2-ylformamido)acetic acid, 2TMS	Regulation of root growth Stress Signalling	Satoh and Nomura, (2017)
15	1,4-Benzenediol, 2,5-bis(1,1-dimethylethyl)- (aka TBHQ-like structure)		

Note: M/Z value: Mass to charge ratio



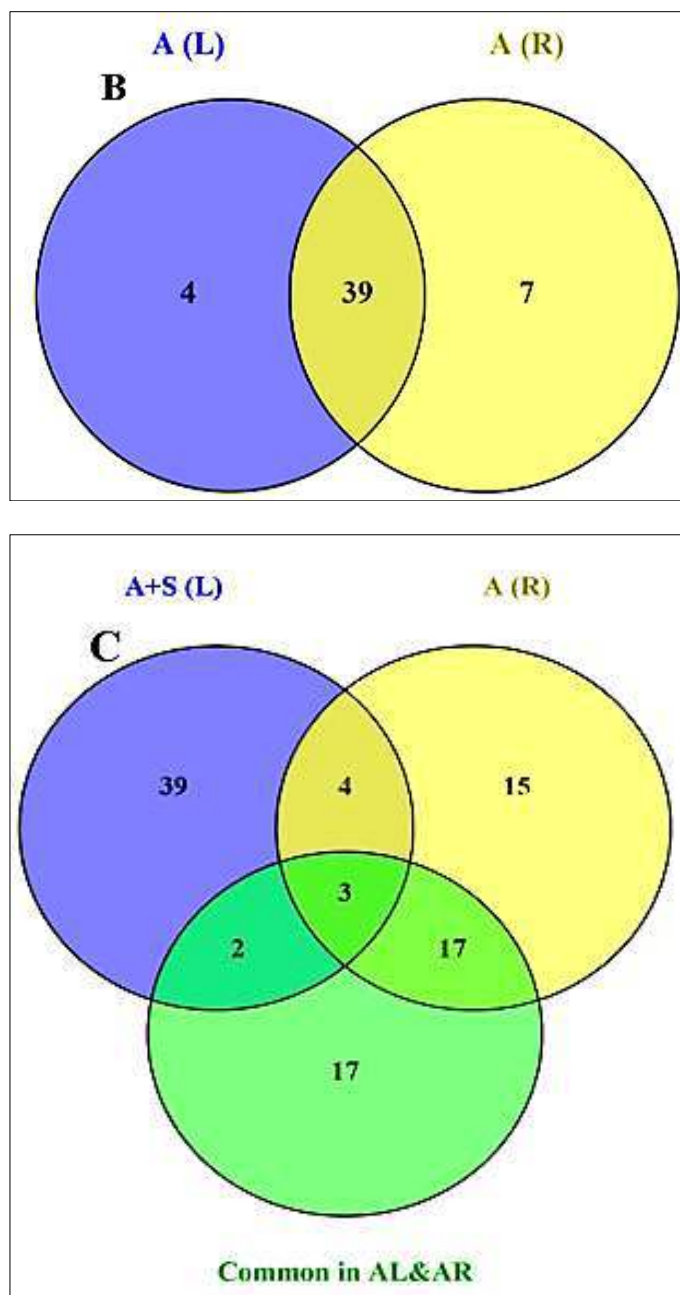


Fig 4: GCMS analysis of root exudates of *Alternanthera* from split root experiment A) Chromatogram of GC-MS analysis of root exudates of A (R) compartment in split root system; B) Venn diagram of comparing the metabolites from A(L) control and A(R) control C) Venn diagram of comparing the metabolites from A + S (L), A (R) and *Alternanthera* alone

Similarly, *Alternanthera* root exudates of a split-root system showed major deviations in metabolism in relation to sandalwood. The two compartments had 39 metabolites that were homogeneous under control conditions (Fig 4B). Comparison of A+S(L) and A(R) and control set showed that only 3 metabolites were shared by the conditions whereas 15 metabolites were only in the right compartment which never was directly exposed to sandalwood (Fig 4C), which has been indicative of systemic signalling. In the distinct metabolites of A(R), the defence and stress-signalling compounds were mainly found, such as 4H-Furo[3, 2 -b]-pyrrole -5 -carboxylic acid, which is tied to plant - microbe signalling. The other compounds that were detected included 2,4-di-tert-butylphenol, which causes lipid peroxidation; 2- (pyridin 2-ylformamido) acetic acid (2TMS), which controls root growth and is a signalling molecule of stress; and antibiotic-related compounds including Madumycin II (Fig 4A).

Identification and Functional Annotation of L-Lysine (KEGG: C00047) in the Split-Root Experiment

KEGG compound annotation of unique compounds found in the right compartment of the three host plants confirm the presence of L-lysine (KEGG ID: C00047) in the exudates of *Casuarina* from right compartment of a Split-Root System Exposed to Sandalwood in the left Compartment. The compound was verified based on molecular properties matching the molecular formula $C_6H_{14}N_2O_2$, with an exact mass of 146.1055 and molecular weight of 146.19 g/mol. Cross-validation of the compound identity was done using KEGG, PubChem (CID: 3349), ChEBI (CHEBI: 18019), and CAS registry number (56-87-1).

Pathway analysis of the enriched pathways revealed that L-lysine is involved in a number of primary and secondary metabolic pathways. It is involved in lysine biosynthesis (map00300) and lysine degradation (map00310), in amino - biosynthesis (map01230) and general metabolism

(map01100). Also, L-lysine is used in synthesizing secondary metabolites (map01110), such as the synthesis of alkaloids (map01064) as well as siderophores (map00975) and plant secondary metabolites (map01060). The module analysis shows that L-lysine is used by various lysine-biosynthetic pathways, which include the succinyl-DAP pathway (M00016), the acetyl-DAP pathway (M00525), and the AAA pathway (M00030), and degradation modules producing saccharopine and acetoacetyl-CoA (M00032) (Table 6). Enzyme mapping indicates that lysine is involved in reactions catalyzed by various enzyme families-aminotransferases (EC 2.6.1.x), oxidoreductases (EC

1.4.x.x), decarboxylases (EC 4.1.1.x), and ligases (EC 6.3.x.x): this reflects its important position in metabolism. The KEGG BRITE hierarchy combined with a functional classification proves that lysine is an amino acid that is common and has broad biological significance. The determination of L-lysine in the samples indicates that there is an active amino acid metabolism and can be associated with protein synthesis, nitrogen processing, and perhaps secondary pathways. Its discovery further indicates that L-lysine is involved in central metabolism pathways and metabolism that are triggered by biotic and abiotic stress.

Table 6: L-lysine is involved metabolic pathways identified through KEGG pathway analysis

KEGG pathway ID	Pathway name
map00300	Lysine biosynthesis
map00310	Lysine degradation
map00470	D-Amino acid metabolism
map00780	Biotin metabolism
map00960	Tropane, piperidine and pyridine alkaloid biosynthesis
map00970	Aminoacyl-tRNA biosynthesis
map00975	Biosynthesis of various siderophores
map00997	Biosynthesis of various other secondary metabolites
map01060	Biosynthesis of plant secondary metabolites
map01064	Biosynthesis of alkaloids derived from ornithine, lysine and nicotinic acid
map01100	Metabolic pathways
map01110	Biosynthesis of secondary metabolites
map01120	Microbial metabolism in diverse environments
map01230	Biosynthesis of amino acids
map02010	ABC transporters

Discussion

The split-root experiment showed that sandalwood association causes a strong and systemic metabolic reprogramming response in both *Mulberry*, *Casuarina* and *Alternanthera*. The large overlap of metabolites between left and right compartments under control conditions confirms that the split-root system did not introduce any variability, and that root exudation was metabolically homogeneous in the absence of sandalwood. The composition of root exudates is relatively stable under controlled hydroponic conditions unless affected by biotic interactions (Bais *et al.*, 2006; Badri and Vivanco, 2009).

In *Mulberry*, the dramatic reduction in shared metabolites (from 37 in the control to 5 in the sandalwood-treated samples) and the appearance of many unique metabolites in both compartments reflect a strong biochemical modulation. Notably, there were also large changes in the right compartment, which was not exposed to sandalwood, and this provides evidence for systemic root-to-root transmission of signals. Systemic transmission of signal between roots separated in space is well established and is typically mediated by vascular transport of hormones such as jasmonic acid, ethylene, auxins, and reactive oxygen species (Karban *et al.*, 2006; Shah and Zeier, 2013). The sole detection of (S)-glyceraldehyde in right compartment indicates a modulation of primary carbon metabolism, which may be indicative of an elevated glycolytic flux or a redirection of carbon to facilitate stress adaptation (Bolton, 2009). Modulation of central carbon metabolism is a well-established response to biotic stress and resource allocation (Caretto *et al.*, 2015).

Casuarina and *Alternanthera* also showed a similar but more drastic metabolic reprogramming. The finding of L-lysine in exudates of *Casuarina* indicates the activation of

amino acid metabolism and possibly the activation of nitrogen remobilization pathways, which are usually activated under stress conditions (Veiga-da-Cunha *et al.*, 2012). The finding of methoxyacetic acid esters and 2-adamantanone oxime also indicates the activation of secondary metabolism and bioactive compound production, which are usually linked to plant-plant or plant-parasite interactions (Inderjit and Duke, 2003)^[13].

The identification and KEGG based functional annotation of L-lysine (C00047) add further mechanistic insight. Lysine is a key player in nitrogen metabolism and is involved in lysine biosynthesis and degradation, amino acid biosynthesis, and secondary metabolite biosynthesis. Amino acids, such as lysine, are used as precursors for the synthesis of alkaloids and other nitrogenous secondary compounds that play a role in defense (Facchini, 2001)^[10]. Lysine degradation is also a source of signaling molecules that play a role in stress adaptation (Arruda *et al.*, 2000)^[12].

Conclusion

The split-root experiment shows that sandalwood (*Santalum album*) causes strong and systemic metabolic reprogramming in host plants even in the absence of direct exposure. Under normal conditions, the metabolite profiles of *Mulberry*, *Casuarina* and *Alternanthera* in the left and right root compartments were very similar, but when sandalwood was introduced into one of the compartments, there was a strong reduction in the shared metabolites and the appearance of a large number of unique metabolites in both the sandalwood-exposed and non-exposed compartments. The identification and metabolic pathway assignment of L-lysine (KEGG C00047) also emphasize the activation of amino acid metabolism and nitrogen redistribution, suggesting the regulation of primary and

secondary metabolism in early host-parasite interaction. The results strongly support the idea that host plants are able to recognize sandalwood-related signals and have the capacity to activate long-distance signaling in the host root system, which respond to metabolic reorganization.

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