

ISOLATION AND CHARACTERIZATION OF *PICHIA OCCIDENTALIS* MHY1: BIOSURFACTANT-PRODUCING YEAST WITH PLANT GROWTH-PROMOTING TRAITS

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Abstract–The objective of this research is to isolate and evaluate biosurfactant-producing yeasts with beneficial traits for plant growth and to assess their impact on seed germination in selected crops. Five yeast isolates (YHA1, YHA2, MHY1, YHA4, YHA5) were recovered using enriched YEPD and MEA media and characterized based on morphological features. Semi-quantitative screening methods, including emulsification index (EI 24%), oil displacement, microplate assay, and Parafilm M test, were employed to evaluate biosurfactant activity. Among the isolates, MHY1 exhibited the highest emulsification index with kerosene, while YHA1 was most effective with engine oil. MHY3 and YHA5 demonstrated the greatest oil displacement (4.5 cm). All isolates were positive in the microplate and Parafilm M assays, but none tested positive for CTAB.PGP trait screening revealed that MHY1 and YHA5 produced indole-3-acetic acid (IAA), with MHY1 also showing moderate siderophore activity. All isolates were negative for phosphate/zinc solubilization and hydrogen cyanide production. Ammonia production varied, with YHA4 and YHA5 being strong producers. Molecular identification via 18S rRNA sequencing confirmed MHY1 as *Pichia occidentalis*. FTIR analysis of MHY1's biosurfactant indicated functional groups characteristic of glycolipids or lipopeptides. Plant growth trials using paddy, pearl millet, and sorghum revealed enhanced germination, seedling vigour, and biomass in seeds treated with MHY1, significantly outperforming controls. MHY1-treated seeds showed the highest seed vigour indices across all crops, suggesting their potential as a plant growth promoter. These findings highlight the dual functionality of MHY1 in biosurfactant production and plant growth promotion, indicating its potential for future use in sustainable agricultural applications.

INTRODUCTION

Industries are gradually transitioning to green alternatives in response to increased environmental concerns and consumer needs for eco-friendly options. Yeast-derived bio surfactants have emerged as a proficient option, offering advantages such as biodegradability, low toxicity, and stability under extreme conditions (Fernandes *et al.*, 2023). These microbial surfactants, particularly from yeast strains like *Candida*, *Rhodotorula*, and *Saccharomyces*, present a viable alternative to synthetic surfactants (Amaral

et al., 2010). Yeast have proven significant potential in promotion of plant growth by mechanisms that include nutrient solubilization, siderophore production along with enhancement of stress tolerance. As components of the rhizosphere microbiome, yeasts are known to interact with plants and contribute to the improvement of plant health and development. Strains such as *Saccharomyces cerevisiae* and *Trichosporon ovoides* have been reported to solubilize inorganic phosphate and produce siderophores, thereby facilitates the uptake of essential nutrients by plants (Fernandes *et al.*,

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2023). Resistance to heavy metals has also been observed in these yeasts, aiding the promotion of plant growth in contaminated soils. Endophytic colonization of plant tissues by yeasts has been associated with pathogen inhibition and the production of beneficial secondary metabolites. Abiotic stress tolerance, including drought and salinity, has been enhanced in plants through yeast interactions, supporting their role in sustainable agriculture. Root exudates released by plants have been shown to attract yeasts, resulting in beneficial associations within the rhizosphere (Chen *et al.*, 2022). These interactions are recognized for their role in the improvement of soil health and nutrient cycling, ultimately leading to higher crop productivity and enhanced plant growth (Bright *et al.*, 2022). The application of yeast-based bioinoculants has been proposed as an eco-friendly strategy to enhance plant resilience against both abiotic and biotic stresses, offering a sustainable alternative to conventional chemical fertilizers (Raish *et al.*, 2025). This study aimed to isolate yeasts from traditionally fermented beverages and characterize the most efficient biosurfactant-producing strain for its potential in promoting plant growth.

MATERIALS AND METHODS

Isolation and morphology characterization of yeast from traditionally fermented beverages (TFBs)

Yeast isolates from TFBs were obtained by spread plating serial dilutions (10^1 to 10^6), prepared by suspending 1 ml of the sample in 9 mL of sterilized water. For each dilution, 0.1 ml was plated on YEPD agar (malt extract 3.0 g/l, yeast extract 3.0 g/l, peptone 5.0 g/l, dextrose 10.0 g/l, and agar 20.0 g/l) supplemented with ampicillin (0.05 g/l) and adjusted pH (5.0) with 1N HCl. The plates were subsequently incubated at 30 °C for 48 hr. Distinct yeast colonies were then transferred to yeast malt agar to isolate pure strains. Colonies that developed on the plates were purified using the streak plate method and examined microscopically for morphology, surface characteristics, and elevation. Screening of yeast-derived bio surfactants

Emulsification activity

The emulsification index (EI_{24}) assay was conducted by following the method demonstrated by Onishi *et al.* (2021). Accordingly, 2 ml of kerosene/engine oil/diesel/olive oil was mixed with 2 ml of cell culture

supernatant in a 1:1 volume ratio. To aid in emulsification, the mixture was vortexed for two minutes. Measurements were taken for both the emulsified layer height and the total height of the liquid column following a 24-hour incubation period at room temperature. The following equation was used to determine the emulsification index (EI):

$$\text{Emulsification Index } 24 \% = \frac{\text{Height of the emulsified layer}}{\text{Total height}} \times 100$$

Oil displacement assay

The oil displacement method was employed to measure the diameter of the clear zone formed when a surfactant-containing solution was applied to the oil-water interface, reflecting the bio surfactant's surface tension-reducing efficiency. A 90 mm diameter Petri plate was filled with 15 ml of distilled water. Then, 100 µl of engine oil was added to the surface, followed by 20 µl of cell culture supernatant. After 30 sec, the clear halo diameter was measured under visible light. SDS served as a positive control, and uninoculated MSM was used as the negative control (Rodrigues *et al.*, 2006).

Microplate assay

Surface activity was qualitatively assessed using the microtiter plate assay for individual strains, as explained by Balakrishnan *et al.* (2022). The culture supernatant (100 µl) was added to wells coated with engine oil in a 96-well microplate. A grid-lined baking sheet was used to observe optical distortion caused by the curvature of the liquid surface. Distortion of the grid image, reflecting changes in surface tension, indicates the presence of biosurfactant

Parafilm-M test

25 µl of culture supernatant was placed onto a Parafilm M strip to assess surface activity on a hydrophobic surface. After 1 minute, the droplet shape was examined. A flattened droplet indicated the presence of biosurfactant, whereas a dome-shaped droplet suggested its absence (Patel, 2020).

Characterization of plant growth-promoting properties in yeast

Indole acetic acid production

Qualitative analysis for indole acetic acid (IAA) production was done by spot inoculating actively growing yeast strains onto nutrient agar enriched with 5/mM L-tryptophan. Plates were maintained at 30/°C for 48/h, after which pre-treated 5/mm

Whatman No. 1 filter discs with Salkowski reagent were placed onto the colonies. The development of a pink coloration indicated positive IAA production (Sun *et al.*, 2014).

Solubilisation of phosphate

Spot inoculation of cultures was done on Pikovskaya agar with tricalcium phosphate as insoluble phosphorous. Plates were then incubated at 30 °C for 72 h. Formation of a zone of clearance around colonies was considered as positive result (da Silva *et al.*, 2022).

Solubilisation of zinc

The Bunt and Roveria medium contain 0.1% zinc oxide (ZnO) as an insoluble zinc source. Plates were incubated at 30 °C for 72 hours. The formation of a zone of clearance around the colonies was deemed a positive result (Bhatt and Maheshwari, 2020).

HCN (Hydrogen cyanide) production

A positive result for HCN production was obtained by spot inoculating actively growing cultures on nutrient agar plates supplemented with glycine (4.4 g/l) and filter paper impregnated with a 0.5% picric acid solution made with 2% sodium carbonate. The cultures were then incubated at 30 °C for 24 to 48 hours. The positive result for HCN generation is shown by a hue shift from orange-red to brown (Bright *et al.*, 2022).

Siderophore qualitative assay

In this qualitative approach, CAS agar plates were prepared by combining 100/ ml of CAS reagent with 900/ml of sterilized LB agar. Each plate is spot inoculated with 4 bacterial strains and include an uninoculated control plate. Incubate the plates at 30 °C for 5-7 days, and then observe for bacterial colonies with an orange color zone indicating siderophore production (Bright *et al.*, 2022)

Ammonia production

Production of Ammonia by all the Yeasts spp. was measured in peptone water and cultured in a shaker incubator for 48 h at 30 °C. Nessler's reagent (0.1 ml) was added to test for ammonium accumulation, and the formation of a yellow-brown colour was monitored (Chen *et al.*, 2022).

Molecular characterisation and construction of phylogenetic tree of potential yeast isolate MHY1

DNA was extracted from the yeast isolate and

evaluated for quality on a 1.0% agarose gel. The 18S rRNA gene was amplified with NS1 and NS4 primers. Then the product was purified and sequenced in both directions using the BDT v3.1 Cycle Sequencing Kit on an ABI 3730xl Genetic Analyzer. The reverse and forward reads were aligned to form a consensus sequence, which was then analysed using BLAST against the NCBI GenBank 'nr' database. Clustal W was used to align the top ten matching sequences, and MEGA 11 was used to create a phylogenetic tree (Archana *et al.*, 2019).

Biosurfactant production by MHY1 strain

Biosurfactant (BS) production by yeast was done using Minimal Salt Medium (MSM) with the composition (g/l): MgSO₄ (0.5), KHPO (1.0), NaNO₃ (1.5), and CaCl₂ (0.002), FeSO₄ (0.01), supplemented with 8% (v/v) soybean oil as the hydrophobic carbon source. Yeast isolate MHY1, pre-cultured in YM broth for 24 hours, was inoculated into MSM at 6% (v/v) and incubated at 30/°C for 7 days under shaking conditions (110/rpm) (Archana *et al.*, 2019).

Extraction of biosurfactant

Yeast cultures were centrifuged for 15 min at 4000 rpm, and after two rinses with distilled water, the cell pellets were retrieved

Solvent Extraction with Chilled Acetone

The solvent extraction method using chilled acetone is commonly employed for the recovery of various bio-compounds, including bio surfactants. Chilled acetone is added to the fermentation broth or solution containing bio surfactants, leading to the precipitation of the target compound due to the low solubility of bio surfactants in cold solvents. This method effectively separates the biosurfactant from the broth by promoting its crystallization or precipitation, which can then be collected by centrifugation or filtration (Rahmalia *et al.*, 2015).

Characterization of biosurfactant by using FT-IR

Fourier transform infrared spectroscopy (FT-IR) was used to characterize biosurfactant extract (FTIR, 400 Perkin Elmer). The signals were collected from 400 to 4000 wave numbers with a resolution of 4 cm⁻¹ (Fernandes *et al.*, 2023).

Assessment of five yeast strains for germination percentage

A seed germination experiment was performed

using Sorghum, Paddy, and Pearl Millet. 2% sodium hypochlorite was used to surface sterilize the seed for 2 min, and then washed 7 times with sterilized distilled water. Following that, the seeds were treated with 70% ethanol for 1 min followed by sterilized distilled water 6 times. Petri plates with blotting paper placed inside were autoclaved at 121 °C for 15 min. Each petri plate contained at least 10 seeds, incubated in dark for five days and evaluated for percentage seed germination using the formula,

$$= \frac{\text{Number of seeds germinated}}{\text{Total number of seeds}} \times 100$$

Assessment of plant growth by the paper towel method using yeast strains

Plant growth of Paddy, Pearl Millet, and Sorghum was analysed by paper towel method as per International Seed Testing agency (ISTA) rules (Association, 1999). Paddy, Pearl Millet, Sorghum seeds were washed thoroughly with sterile distilled water and surface sterilized as described in the previous step. These surface-sterilised seeds were further treated with five different yeast strains combined with 1% CMC (Carboxy Methyl Cellulose) separately for 1h. Seeds treated with sterile distilled water containing CMC are considered the control. All the seeds with respective treatments and controls were rolled in a paper towel and incubated for 8 days at 25 °C. Parameters like root length, shoot length and dry plant weight, and wet plant weight were recorded at the end of incubation (Bright *et al.*, 2022).

RESULTS

Isolation of yeasts

All samples were processed into the enriched media of YEPD amended with malt and MEA to identify the biosurfactant producers and a total of 5 yeasts were isolated (Fig. 1). Morphological characteristics such as colony colour, surface, elevation and margin were recorded (Table 1). Subsequently, 5 Yeast isolates were selected based on their morphological

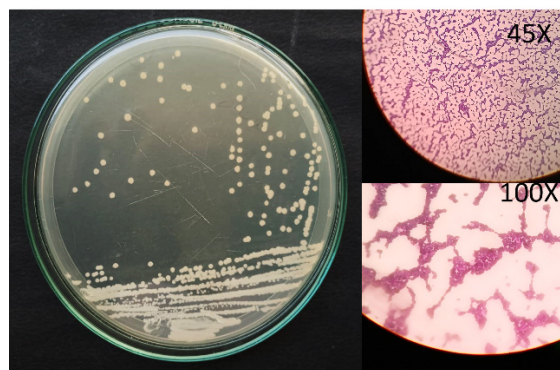


Fig. 1. Pure culture of yeast spp. and microscopic image under 45x and 100x

characteristics.

Morphological characterization of yeasts

Screening for biosurfactant production

All 5 yeast isolates were tested for several screening tests for biosurfactant activity.

The emulsification activity of different yeast strains with various hydrocarbons reveals distinct patterns. YHA1 shows moderate emulsification visually with engine oil and diesel, with the graph indicating highest activity with engine oil (~85%), followed by diesel and olive oil, but weak with kerosene. YHA2 demonstrates a strong visible emulsion layer with engine oil, confirmed by high emulsification activity (~90%) in the graph, and moderate activity with diesel and olive oil, but again, weak with kerosene. MHY1 exhibits moderate emulsification with engine oil and diesel, with the graph showing engine oil as the most emulsified (~75%), while other hydrocarbons show lower activity. YHA4 displayed strong emulsification with engine oil (~95%) and diesel (~60%) activity, with kerosene showing the lowest. YHA5 stands out with the most consistent emulsification across all hydrocarbons, and the graph reflects this with the highest emulsification activity for engine oil (~100%) and significant activity for both diesel and olive oil (Fig. 2).

Table 1. Morphological and colony characteristics of yeasts

Isolates	Colony Colour	Surface	Elevation	Margin
YHA1	Circular White	Smooth	Flat	Curled
MHY1	Circular White	Smooth	Convex	Curled
YHA2	Circular White	Smooth	Convex	Curled
YHA4	Glistening	Rough	Umbonate	Lobate
YHA5	Glistening	Rough	Raised	Undulate

Oil displacement assay

The oil displacement assay is a sensitive and simple approach that requires little amount of culture supernatant. Five isolates had oil displacement ranged from 1 to 4.5 cm in diameter, isolates MHY3 and YHA5 having the highest oil displacement capability (4.5cm) (Fig. 3).

Microplate assay

The optical irregularity of the microplate assay is used to carry out the test. In a hydrophobic well, pure water has a flat surface, but the surfactant containing fluid has a concave surface that becomes uneven. For microplate assay all five isolates were positive (Fig. 4).

Parafilm- M Test

The Parafilm M test is an effective preliminary screening tool for assessing biosurfactant

production in microorganisms. Here all five yeast isolates, sample 1 - YHA1, sample 2 - YHA2, sample 3 - MHY1, sample 4 - YHA4, and sample 5 - YHA5, exhibited positive results for the test (Fig. 5).

Screening for plant growth-promoting yeast (PGPY) traits

All five yeast isolates were evaluated for plant growth-promoting (PGP) characteristics, such as phosphate and zinc solubilization, indole-3-acetic acid (IAA) production, and siderophore production. Among them, isolates MHY1 and YHA5 tested positive for IAA production, indicated by the appearance of a pink coloration on paper discs following the application of Salkowski reagent. In contrast, all isolates tested negative for phosphate and zinc solubilization, as no clear zones were observed in the respective assays. Among yeast isolates, MHY1 showed siderophore

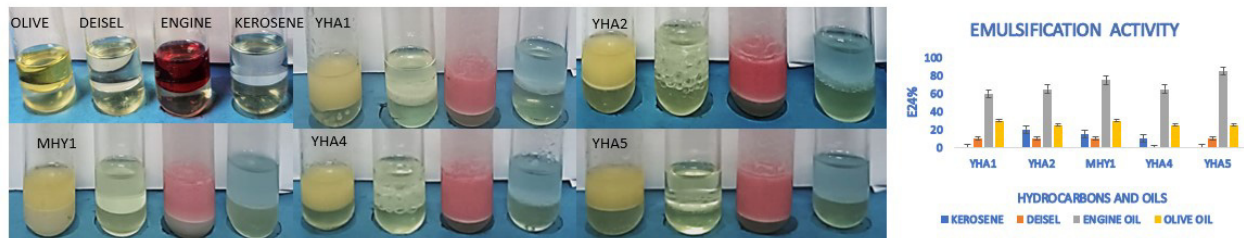


Fig. 2. Emulsification activity of different yeast strains with various hydrocarbons and oils by graphical representation.

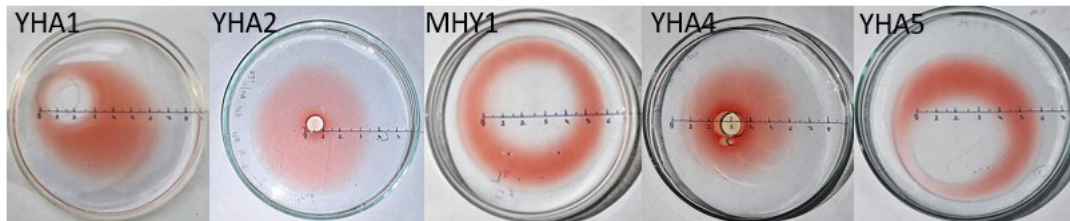


Fig. 3. Oil displacement assay of different yeast isolates



Fig. 4. Microplate assay of different yeast isolates

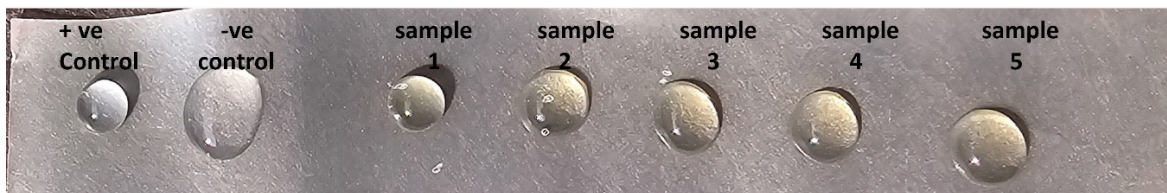


Fig. 5. Parafilm-M test for yeast isolates

Further, all isolates showed negative for the HCN production as there was no change in colour of the filter paper after adding the picric acid. Isolates YHA4 and YHA5 showed positive results for ammonia production with a varying intensity of yellow-brown colour upon addition of Nessler's reagent. The isolates YHA1, YHA2, and MHY1 were classified as weak producers based on the ammonia production exhibited a change of the colour to dark yellow and light yellow, respectively (Fig. 6).

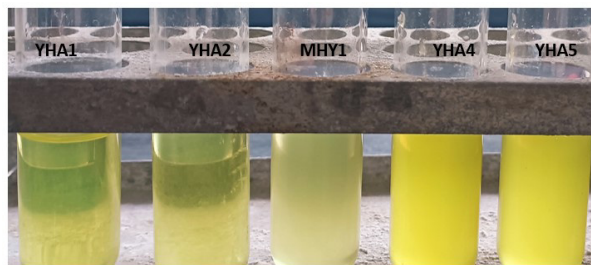


Fig. 6. Production of ammonia by yeast isolates

Gene sequencing and phylogenetic analysis of efficient strain

MHY1 is identified using 18S rRNA. BLAST was performed on ITS region sequence with NCBI GenBank database. The resemblance of the sequence to the species was analysed by NCBI BLAST. Sample which was labelled as MHY1 showed high similarity with *Pichia occidentalis*, submitted to the NCBI, and accession numbers were assigned. The

first ten sequences were chosen based on the maximum identity score, and were aligned by using multiple alignment software program Clustal W. Phylogenetic tree was generated using neighbour joining method MEGA X software and dot matrix used to show distance matrix (Fig. 7).

FT-IR spectroscopy analysis

The FTIR was analysed to determine the functional groups in the biosurfactant compound (Fig. 8). O-H stretching vibrations were identified as the cause of a broad absorption band observed at about 3375 cm^{-1} , that suggested the existence of hydroxyl groups, which are characteristic of alcohols or phenols. Peaks near 3010 , 2925 , and $2854/\text{cm}^{-1}$ were attributed to aliphatic chains showing C-H stretching vibrations, indicating the presence of -CH and -CH₂ groups typically found in fatty acid moieties. A sharp peak at $1743/\text{cm}^{-1}$ was evidence of C=O stretching vibrations, commonly associated with ester or carboxylic acid functional groups. The band at $1644/\text{cm}^{-1}$ could be assigned to C=C stretching of alkenes or possibly amide groups. Additional peaks at 1461 , 1371 , and $1231/\text{cm}^{-1}$ were associated with bending vibrations of -CH₃ and -CH₂ groups and C-O stretching, respectively. The region between 1098 and $624/\text{cm}^{-1}$ showed multiple peaks, which were linked to C-O-C and C-H bending vibrations, suggesting the presence of polysaccharide or glycosidic components. These

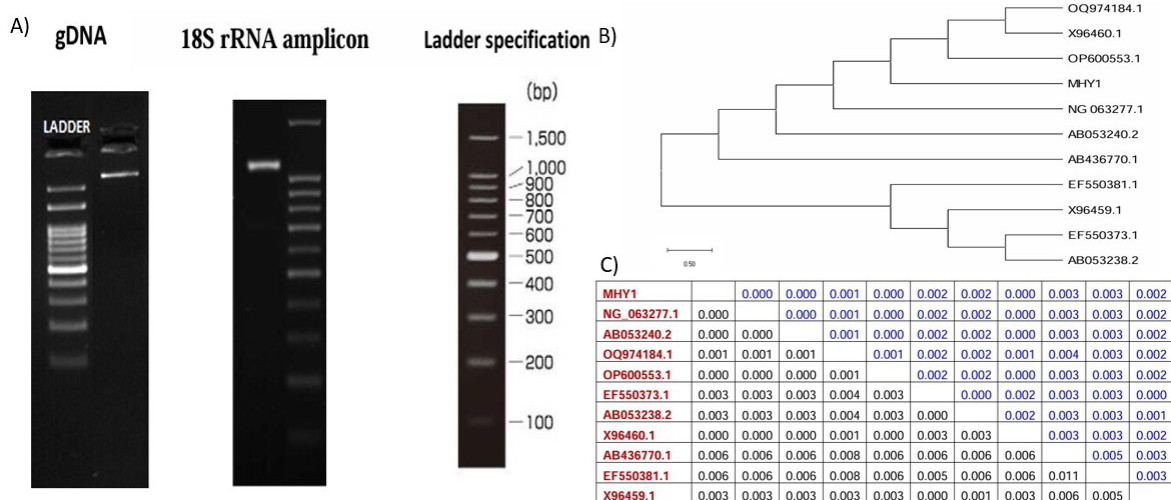


Fig. 7. Molecular identification and phylogenetic analysis of the MHY1 *Pichia occidentalis* yeast isolate based on 18S rRNA gene sequencing. A) Agarose gel electrophoresis showing genomic DNA (gDNA) extraction, successful amplification of 18S rRNA gene, and the DNA ladder used for size estimation. B) Phylogenetic tree was constructed using the Neighbor-Joining method indicates the link between MHY1 and closely related yeast strains based on 18S rRNA gene sequences. C) Pairwise genetic distance matrix illustrating the nucleotide divergence between MHY1 and reference strains, with lower values indicating higher sequence similarity.

findings collectively indicated that the biosurfactant produced may contain a glycolipid or lipopeptide structure, comprising lipid chains along with hydroxyl and carbonyl functional groups.

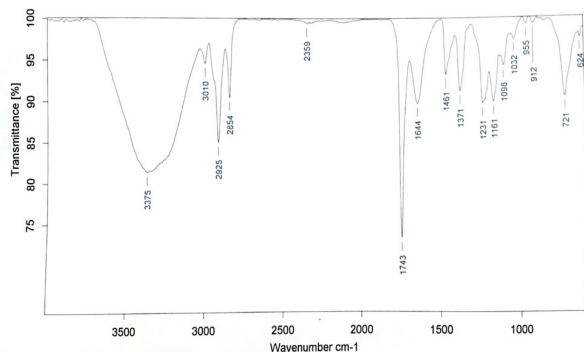


Fig. 8. FTIR Analysis of biosurfactant production by MHY1 strain

Evaluation of yeast strains for plant growth promotion

The evaluation of five yeast strains (MHY1, YHA1, YHA2, YHA4, and YHA5) for plant growth promotion using the paper towel method over 15 days (Fig. 9) revealed that MHY1 was the most effective across all tested crops-paddy, pearl millet, and sorghum. In paddy, MHY1 resulted in 100% germination, a shoot height of 5.21 cm, root height of 3.69 cm, dry weight of 0.41 g, and the highest seed vigour index of 889, significantly outperforming the

control (2.42 cm shoot, 4.12 cm root, 0.34 g dry weight, and 654 vigour index). For pearl millet, MHY1 also achieved 100% germination with shoot and root heights of 4.37 cm and 3.06 cm, respectively, a dry weight of 0.67 g, and the highest seed vigour index of 743, while the control only had 50% germination and a vigour index of 175.5. In sorghum, MHY1 showed 80% germination, shoot and root heights of 4.15 cm and 4.55 cm, dry weight of 0.38 g, and a seed vigour index of 696, compared to the control's 50% germination and 191.5 vigour index. YHA1 showed moderate results (e.g., paddy vigour index: 768; pearl millet: 524.8), while YHA4 and YHA5 had variable improvements over the control. In contrast, YHA2 exhibited the weakest



Fig. 9. Evaluating yeast strains for plant growth promotion by the paper towel method

Table 2. Seed germination assays of paddy, pearl millet, and sorghum following treatment with the yeast strains

Samples	Crops	Germination Rate (%)	Shoot height (cm)	Root height (cm)	Dry weight (g)	Seed vigour index
Control	Paddy	100%	2.42 ± 0.02	4.12 ± 0.03	0.34 ± 0.02	654
	Pearl Millet	50%	1.83 ± 0.01	1.68 ± 0.02	0.09 ± 0.01	175.5
	Sorghum	50%	2.26 ± 0.02	1.57 ± 0.03	0.28 ± 0.03	191.5
MHY1	Paddy	100%	5.21 ± 0.12	3.69 ± 0.01	0.41 ± 0.01	889
	Pearl Millet	100%	4.37 ± 0.01	3.06 ± 0.04	0.67 ± 0.02	743
	Sorghum	80%	4.15 ± 0.03	4.55 ± 0.01	0.38 ± 0.02	696
YHA1	Paddy	80%	4.56 ± 0.01	5.05 ± 0.02	0.24 ± 0.01	768
	Pearl Millet	80%	3.94 ± 0.02	2.62 ± 0.04	0.51 ± 0.01	524.8
	Sorghum	70%	2.94 ± 0.03	2.37 ± 0.01	0.35 ± 0.05	231.7
YHA2	Paddy	40%	2.63 ± 0.01	1.46 ± 0.02	0.24 ± 0.02	162.4
	Pearl Millet	60%	3.04 ± 0.13	1.75 ± 0.01	0.25 ± 0.01	287.4
	Sorghum	60%	2.52 ± 0.01	1.98 ± 0.01	0.30 ± 0.03	270
YHA4	Paddy	90%	3.35 ± 0.21	3.11 ± 0.02	0.38 ± 0.04	581.4
	Pearl Millet	70%	3.50 ± 0.04	2.25 ± 0.01	0.39 ± 0.01	402.5
	Sorghum	60%	2.43 ± 0.03	1.67 ± 0.19	0.30 ± 0.04	246
YHA5	Paddy	80%	3.45 ± 0.03	3.72 ± 0.02	0.41 ± 0.04	573.6
	Pearl Millet	60%	3.14 ± 0.17	1.82 ± 0.05	0.37 ± 0.02	297.6
	Sorghum	50%	2.51 ± 0.31	1.84 ± 0.02	0.33 ± 0.01	217.5

performance, particularly in paddy with only 40% germination and the lowest vigour index of 162.4 (Table 2).

DISCUSSION

Despite the promising findings regarding yeast-derived bio surfactants, several challenges hinder their widespread application. The scalability of production processes remains a significant barrier, as many studies are conducted at a laboratory scale without subsequent industrial validation. Additionally, the economic feasibility of biosurfactant production compared to conventional surfactants requires further investigation. Yeast-derived bio surfactants have emerged as attractive alternatives to synthetic surfactants because of biodegradability, low toxicity, and environmental compatibility. These surface-active substances, yielded by yeasts such as *Candida bombicola*, offer diverse applications across multiple industries, including pharmaceuticals, agriculture, and environmental remediation (Fernandes *et al.*, 2023; Sarubbo *et al.*, 2022). MHY1 consistently outperformed all other isolates and the control in enhancing seed germination and early seedling growth across paddy, pearl millet, and sorghum, establishing it as a strong candidate for use as plant growth-promoting yeast (PGPY). In contrast, YHA1 and YHA4 demonstrated moderate effectiveness, particularly in paddy and pearl millet, but were less effective in promoting sorghum growth. YHA2 exhibited the weakest performance across all crops, due to the possible absence of key growth-promoting traits or the presence of inhibitory effects under the experimental conditions. The Present work highlights the broader implications of integrating these yeast isolates into agricultural practices. Overall, the study findings demonstrate that the MHY1 strain (*Pichia occidentalis*) exhibits a dual functionality as an efficient biosurfactant producer, along with an effective plant growth promoter. By enhancing soil fertility, promoting plant growth, and providing natural defence mechanisms against pathogens, these yeasts offer a promising alternative to chemical inputs. The research advocates for the adoption of these microorganisms as key components of sustainable farming systems, which could lead to increased agricultural productivity while minimizing environmental impact (Ruspi *et al.*, 2024).

CONCLUSION

Yeast isolates were first isolated and screened for biosurfactant production. They were further evaluated for plant growth-promoting (PGP) traits. Among these isolates, the most efficient strain, MHY1, was selected for gene sequencing and identified as *Pichia occidentalis*. Among the tested isolates, MHY1 demonstrated the most promising plant growth-promoting traits, marked improvement in seed germination, seedling growth parameters, and vigour indices. YHA2 exhibited the weakest performance across all crops, indicating a possible absence of key growth-promoting traits or the presence of inhibitory effects under the experimental conditions. These findings suggest its potential utility in sustainable agriculture, particularly for enhancing the early growth of cereal crops like paddy, pearl millet, and sorghum. Other isolates, such as YHA1 and YHA4, showed moderate effectiveness and may also be considered for further evaluation and development as plant growth promoters.

Conflict of Interest

There is no conflict of interest between the authors; all authors contributed directly to the article.

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Author contributions

PGSM Conceived Conceptualization, Methodology, Investigation, Writing – the original draft of manuscript, Writing – review and editing; PGSM and APA had done Formal and Statistical analysis, Software, Visualization, Data curation; BH supervised the work, Project administration, Conceptualization, Methodology, Supervision, Validated and edited the manuscript. All authors, PGSM, APA, and BH, reviewed and approved the final manuscript.

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