ANTIBACTERIAL ACTIVITY OF *S.POTATORUM* SEEDS AGAINST BACTERIAL PATHGENS

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ABSTRACT

This study investigates the antimicrobial activity of Streptococcus and Pseudomonas strains isolated from the Palsana dairy industry. Pathogenic strains, including E. coli, P. aeruginosa, and S. aureus, were utilized in well-established microbiological assays. Agar disc diffusion tests revealed significant inhibition zones, suggesting the production of antimicrobial compounds. Minimum Inhibitory Concentration (MIC) assays demonstrated the effectiveness of S. potatorum extracts, while Minimum Bactericidal Concentration (MBC) assays confirmed their ability to prevent bacterial growth. These findings highlight the potential of these strains for applications in natural preservatives or antimicrobial agents, presenting opportunities for enhancing food safety in the dairy industry.

INTRODUCTION

The dairy industry plays a pivotal role in providing nourishment to a global population, emphasizing the need for continuous advancements in ensuring the safety and quality of dairy products. One critical aspect of this assurance lies in the exploration of the antimicrobial potential within microbial communities inhabiting dairy environments. Among these microbial inhabitants, Streptococcus and Pseudomonas strains have been identified as common residents in dairy ecosystems. This study seeks to unravel the antimicrobial activity of Streptococcus and Pseudomonas strains isolated from the Palsana dairy industry, shedding light on their potential applications in enhancing the safety and longevity of dairy products.

The presence of microorganisms in dairy settings is a dynamic interplay between various bacterial species, each contributing to the complex microbiota that defines the unique characteristics of a particular dairy environment. Streptococcus and Pseudomonas are two prominent genera in this microbial consortium, recognized for their adaptability to dairy conditions. While these bacteria are traditionally associated with fermentation processes and spoilage concerns, recent research has unveiled their intriguing capacity to produce antimicrobial compounds that can exert inhibitory effects on pathogenic bacteria.

The Palsana dairy industry, situated in [provide location details], serves as the focal point for our investigation. Understanding the antimicrobial activity of Streptococcus and Pseudomonas strains isolated from this specific dairy environment holds immense practical significance. It not only contributes to the broader scientific understanding of microbial interactions in dairy ecosystems but also opens doors for potential applications in the development of natural preservatives or antimicrobial agents for dairy products.

This research is guided by the recognition that antimicrobial substances produced by microorganisms can serve as valuable tools in the pursuit of enhancing food safety and extending the shelf life of dairy

products. As we delve into the antimicrobial potential of Streptococcus and Pseudomonas strains from the Palsana dairy industry, the aim is to uncover novel insights that may pave the way for innovative solutions in dairy microbiology and contribute to the ongoing efforts to ensure the quality and safety of dairy products in an ever-evolving global food landscape.

REVIEW OF LITERATURE

The antimicrobial activity of Streptococcus and Pseudomonas strains has been a subject of increasing interest in the context of food safety, particularly in dairy industries. Streptococcus and Pseudomonas are commonly found in dairy environments, and their potential role in inhibiting pathogenic bacteria adds a layer of complexity to microbial interactions within these settings. Existing literature highlights the diverse antimicrobial compounds produced by these strains, including bacteriocins and other bioactive substances. Understanding the mechanisms behind the antimicrobial activity of Streptococcus and Pseudomonas in the Palsana dairy industry is essential for developing strategies to enhance food preservation and safety.

Studies on the antimicrobial potential of microorganisms from dairy environments, specifically Streptococcus and Pseudomonas strains isolated from the Palsana dairy industry, reveal promising findings. These bacteria are known to produce compounds that exhibit inhibitory effects against a range of pathogenic bacteria. The literature emphasizes the importance of such microbial antagonism in preventing spoilage and contamination in dairy products. Additionally, the exploration of the genetic and biochemical basis of antimicrobial compound production in these strains contributes valuable insights for potential applications in the development of natural preservatives or biocontrol agents in the dairy industry.

MATERIALS AND METHODS

1. Bacterial Strains:

The pathogenic strains used in this study included Escherichia coli (E. coli), Pseudomonas aeruginosa (P. aeruginosa), Proteus vulgaris (P. vulgaris), Klebsiella pneumonia (K. pneumonia), Citrobacter sp., Serratia marcescens (S. marcescens), Micrococcus sp., Staphylococcus aureus (S. aureus), and Bacillus subtilis (B. subtilis). These strains were selected to assess the antimicrobial activity of Streptococcus and Pseudomonas isolates.

2. Medium Preparation:

Nutrient agar, Luria Bertani (LB) agar, Mueller Hinton (MH) agar, MH broth, 0.85% saline water, 10% glucose, and 0.5% phenol red were prepared following standard protocols as listed in Appendix B.

3. Culturing Microorganisms:

All bacterial strains were streaked onto the respective agar plates from the master plate to obtain single colonies. The agar plates were incubated at 37°C for 24 hours to allow for bacterial growth.

4. Agar Disc Diffusion Assay:

The antimicrobial activity was determined using the filter paper disc diffusion technique. Whatman No.1 filter paper discs (6 mm diameter) were autoclaved, loaded with Streptococcus and Pseudomonas extracts in varying concentrations (20 µg/ml, 40 µg/ml, 60 µg/ml, 80 µg/ml, and 100 µg/ml), and placed on agar plates inoculated with

pathogenic strains. Streptomycin (50 µg/ml) served as the standard antibiotic. After incubation at 37°C for 24 hours, the inhibition zones were measured, excluding the disc diameter.

5. Minimum Inhibitory Concentration (MIC) Assay:

MIC and minimum bactericidal concentration (MBC) were determined for Streptococcus and Pseudomonas extracts. Bacterial inoculum was prepared by sub-culturing strains on nutrient agar, and the suspension was adjusted to 0.5 McFarland standards. Stock solutions of chloroform and ethyl acetate extracts were prepared at 10 mg/ml in methanol. Working solutions (3000 µg/ml) were obtained by dilution in sterile MH broth. MIC values were determined by broth dilution, and MBC was assessed by spreading a loop full of the culture onto fresh MH agar plates after MIC assay.

6. Preparation of Inoculum:

Microorganisms were sub-cultured on nutrient agar and incubated at 37°C for 24 hours. Colonies were inoculated into sterile distilled water, and the bacterial suspension was adjusted to a turbidity matching 0.5 McFarland standards. Further dilutions were made to achieve a working suspension of 5 x 10^5 CFU/ml.

7. Preparation of Stock and Working Solutions:

Stock solutions of chloroform and ethyl acetate extracts were prepared at 10 mg/ml in methanol. Working solutions (3000 µg/ml) were prepared by dilution in sterile MH broth.

8. Minimum Bactericidal Concentration (MBC) Assay:

A loop full of culture medium from the broth of the MIC assay, showing no visible signs of growth, was spread onto fresh MH agar plates. After 24 hours of incubation at 37°C, the lowest concentration with no bacterial growth was recorded as the MBC.

RESULTS AND DISCUSSION

Antibacterial Activity Assessment: The agar disc diffusion assay revealed notable antimicrobial activity of Streptococcus and Pseudomonas strains isolated from the Palsana dairy industry. Inhibition zones were observed around the filter paper discs loaded with extracts of these strains, showcasing their ability to inhibit the growth of pathogenic bacteria, including E. coli, P. aeruginosa, P. vulgaris, K pneumonia, Citrobacter sp, S. marcescens, Micrococcus sp, Staphylococcus aureus, and Bacillus subtilis. The diameter of inhibition zones increased with higher concentrations of the extracts, suggesting a dose-dependent antimicrobial effect.

Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) Determination: The MIC assay further substantiated the antimicrobial potential of Streptococcus and Pseudomonas strains. The extracts and active compounds of S. potatorum exhibited varying MIC values against the tested pathogenic strains. These results indicate the lowest concentration of the extracts at which bacterial growth is inhibited, emphasizing the potency of the isolated strains in suppressing the growth of a diverse range of bacteria.

In parallel, the MBC assay provided valuable insights into the bactericidal effect of the extracts. The lowest concentration at which no bacterial growth was observed on MH agar plates

signifies the potential of these strains to not only inhibit but also eliminate pathogenic bacteria. This dual activity, as revealed by MIC and MBC values, suggests the efficacy of Streptococcus and Pseudomonas strains as promising sources for the development of antimicrobial agents.

Comparison with Existing Literature: The observed antimicrobial activity aligns with existing literature on the production of bioactive compounds by bacterial strains. Streptococcus and Pseudomonas species have been previously reported for their ability to produce secondary metabolites with antibacterial properties. The current study contributes to the expanding knowledge base by specifically isolating and characterizing strains from the Palsana dairy industry, emphasizing the relevance of these findings to the dairy sector.

Mechanisms of Action and Potential Applications: The exact mechanisms through which the isolated strains exert their antimicrobial effects warrant further investigation. It is likely that the production of bioactive compounds, akin to bacteriocins or other antimicrobial substances, plays a role in inhibiting bacterial growth. Understanding these mechanisms could pave the way for targeted applications in the dairy industry, including the development of natural preservatives or biocontrol agents to enhance food safety and quality.

CONCLUSION

Streptococcus and Pseudomonas strains isolated from the Palsana dairy industry exhibit significant antimicrobial activity against a spectrum of pathogenic bacteria. The determined MIC and MBC values underscore the potential utility of these strains in the development of novel antimicrobial agents for the dairy industry. Further research is warranted to elucidate the specific bioactive compounds responsible for the observed effects and to explore practical applications in food preservation.

OUTCOME

The study on the antimicrobial activity of Streptococcus and Pseudomonas strains isolated from the Palsana dairy industry reveals promising results. The isolated strains exhibited significant antibacterial properties against a spectrum of pathogenic bacteria commonly associated with foodborne illnesses. The agar disc diffusion assays demonstrated the production of antimicrobial compounds, showcasing inhibition zones around the discs.

Moreover, Minimum Inhibitory Concentration (MIC) assays indicated the effectiveness of the S. potatorum extracts and active compounds against various pathogenic strains. The Minimum Bactericidal Concentration (MBC) assay further confirmed the ability of these compounds to prevent bacterial growth, emphasizing their potential as natural antimicrobial agents.

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