



Evaluation of the Anti-Microbial Activity of Zea Mays Seeds

E. Rajani*, SK. Shabana, T. Harika, P. Meghana, K. Saipravallika, CH. Badrinath

Ratnam Institute of Pharmacy, Pidhapolur (V), Muthukur (M), Nellore (Dt), Andhra Pradesh-524343, India

ABSTRACT

The evaluation of maize extract as an antimicrobial agent has yielded promising results, showcasing its efficacy against a spectrum of bacterial strains, encompassing both Gram-positive and Gram-negative types. This efficacy is attributed to the presence of bioactive compounds within maize extracts, including phenols, flavonoids, and terpenoids. Laboratory tests, such as disc diffusion and broth dilution assays, have underscored the substantial antibacterial activity of maize extracts, evidenced by notable zones of inhibition and reductions in bacterial growth. Despite these encouraging findings, further research is indispensable to comprehensively grasp the antibacterial properties of maize extract and its practical applications. A thorough analysis of active compounds, their mechanisms of action, and optimal extraction methods is imperative for translating these findings into tangible benefits. Additionally, safety evaluations and toxicity assessments are paramount to ensure the suitability of maize extract for human and environmental use. With sustained investigation, maize extract stands poised as a sustainable and natural alternative to conventional antibacterial agents, potentially mitigating antibiotic resistance and fostering eco-conscious practices across diverse industries.

Keywords: Gram-positive, maize, antibiotic, environmental, antibacterial agents

ARTICLE INFO

*Corresponding Author:

E. Rajani
Ratnam Institute of Pharmacy, Pidhapolur (V),
Muthukur (M), Nellore (Dt),
Andhra Pradesh- 524343, India.

Article History:

Received 24 Feb 2024
Revised 10 Mar 2024
Accepted 25 Mar 2024
Published 30 April 2024

Copyright© 2024 The Contribution will be made Open Access under the terms of the Creative Commons Attribution-NonCommercial License (CC BY-NC) (<http://creativecommons.org/licenses/by-nc/4.0>) which permits use, distribution and reproduction in any medium, provided that the Contribution is properly cited and is not used for commercial purposes.

Citation: E. Rajani, Evaluation of the Anti-Microbial Activity of Zea Mays Seeds, 2024,11(1): 17-21.

CONTENTS

1. Introduction.....	17
2. Methodology.....	18
3. Results and Discussion.....	19
4. Conclusion.....	20
5. References.....	20

1. Introduction

Medicinal plants harbor remedies for a plethora of diseases, possessing attributes such as antibacterial, antifungal, antioxidant, and anticancer activities. Utilization of plant-derived products, metabolites, or plant-mediated biosynthesized nanoparticles is widespread. Corn (*Zea mays* L), among staple crops like wheat and rice, stands as one of the most extensively cultivated cereal crops globally. Corn silk, the stigma and style of the corn fruit, constitutes a readily available waste material post-harvest [9]. Medically, corn silk (CS), composed of yellowish thread-like strands from *Zea mays* L. (Gramineae) female flowers, finds application in treating various ailments, including irritation and inflammation within the urinary system [12].

Noteworthy benefits of CS include its reported efficacy in reducing hyperglycemia and inhibiting IgE formation by glycoproteins [13,14]. The overuse of antibiotics against infectious diseases has led to the emergence of resistant organisms and adverse side effects, prompting scientists to explore alternative sources, notably medicinal plants, for antimicrobial substances to combat uncommon infections [15]. Consequently, research into the antimicrobial properties of different plant species across various geographical regions has been conducted to develop antibiotics [16]. This study aims to assess the antimicrobial activities of ethanol extract from corn silk of *Zea mays* L

against UTI bacteria and certain food spoilage-causing fungi.

2. Methodology

Chemicals & Reagents

Maize seeds, ethanol (or other suitable solvents), Soxhlet extractor, condenser, round-bottom flask, heating mantle or hot plate, distillation apparatus, filter paper, and optionally, a rotary evaporator for solvent removal.

Method:

To initiate the extraction process, begin by thoroughly cleaning and drying the maize seeds to eliminate any impurities. Next, grind the seeds into a fine powder using an appropriate grinder. Once the seeds are prepared, set up the Soxhlet extraction apparatus, comprising a round-bottom flask at the base, a sample thimble containing the powdered maize seeds, and a condenser positioned at the top. Following the setup, select a suitable solvent such as ethanol, depending on the specific compounds targeted for extraction. Commence the extraction by heating the solvent, causing it to vaporize and ascend through the apparatus. As the vapor condenses in the condenser and drips onto the powdered seeds within the thimble, it dissolves the compounds from the seeds and transports them back to the flask. Allow the extraction process to continue for several hours, typically spanning 6 to 8 hours, to ensure comprehensive extraction of the desired compounds.

Phytochemical Screening

Alkaloids (Mayer's Test):

Mix the plant extract with Mayer's reagent (potassium mercuric iodide). A yellow or cream precipitate indicates the presence of alkaloids. The endpoint is the formation of a yellow or cream-colored precipitate.

Flavonoids (Shinoda Test):

Add magnesium ribbon and a few drops of concentrated hydrochloric acid to the plant extract. A pink or red coloration indicates the presence of flavonoids. The endpoint is the development of a pink or red color.

Tannins (Ferric Chloride Test):

Mix the plant extract with a few drops of ferric chloride solution. A dark green or blue-black coloration indicates the presence of tannins. The endpoint is the formation of a dark green or blue-black color.

Saponins (Froth Test):

Shake the plant extract vigorously with water. The formation of a stable froth indicates the presence of saponins. The endpoint is persistent frothing.

Glycosides (Legal's Test):

Treat the plant extract with glacial acetic acid and add a few drops of ferric chloride. A blue or green coloration indicates the presence of glycosides. The endpoint is the development of a blue or green color.

Steroids/Triterpenoids (Liebermann-Burchard Test):

Mix the plant extract with acetic anhydride, followed by concentrated sulfuric acid. A color change from violet to blue or green indicates the presence of steroids/triterpenoids. The endpoint is a violet to blue or green color change.

Terpenoids (Salkowski Test):

Mix the plant extract with chloroform and concentrated sulfuric acid. A reddish-brown coloration at the interface indicates the presence of terpenoids. The endpoint is the formation of a reddish-brown color at the interface.

Phenols (Ferric Chloride Test):

Mix the plant extract with a few drops of ferric chloride solution. The development of a bluish-black color indicates the presence of phenols. The endpoint is the formation of a bluish-black color.

In vitro Evaluation of Anti-bacterial activity:

Preparation of Microbial Cultures:

To start, *Escherichia coli* species cultures are either obtained from a reliable source or isolated from affected individuals. These microorganisms are then inoculated onto nutrient agar or Sabouraud agar plates.

Procedure:

- **Preparation of Agar Plates:** The appropriate agar medium, either Sabouraud Dextrose Agar (SDA) or modified Dixon's agar, is prepared according to the manufacturer's instructions. This involves weighing, mixing, heating, sterilization by autoclaving, pouring into plates or tubes, and finally, storage.
- **PH Adjustment:** The pH of Sabouraud Dextrose Agar is usually around 5.6, which is suitable for fungal growth without the need for adjustment.
- **Inoculation:** Samples are collected from the affected area using sterile swabs or scraping tools. These samples are streaked onto the surface of agar plates using sterile inoculation loops to ensure even distribution.
- **Incubation:** The inoculated agar plates are then placed in an incubator set to 32-37°C (89.6-98.6°F) for up to 7-14 days, as *Escherichia coli* species typically grow slowly.
- **Observation and Subculturing:** Plates are regularly checked for the presence of yeast-like colonies, which are typically cream to yellowish in color. If colonies appear, they are subcultured onto fresh agar plates to obtain pure cultures.
- **Identification:** Additional tests, such as microscopy, biochemical tests, or molecular methods, are performed to confirm the identity of the isolated *Escherichia coli* species.
- **PH Control (Optional):** The pH of the agar can be checked using pH indicator paper, with *Escherichia coli* species preferring slightly acidic conditions.
- **Storage:** Pure cultures are stored in the refrigerator or frozen for long-term storage.

Determination of the Zone of Inhibition by Agar Diffusion Method:

Procedure:

- **Preparation of Nutrient Agar Plates:** Petri dishes are sterilized and filled with sterile nutrient agar to create a solid medium.
- **Inoculation of Microorganisms:** Bacterial or fungal cultures are streaked or spread onto the surface of

the agar plates using sterile swabs, ensuring even distribution.

- Placement of Discs: Sterile filter paper discs are placed on the surface of the inoculated agar plates, and the substances to be tested are applied onto these discs.
- Incubation: The plates are then incubated at the appropriate temperature for the microorganism being tested.
- Observation: After incubation, the plates are observed for the presence of clear zones of inhibition around the discs, indicating the inhibition of microbial growth.
- Measurement of Zones of Inhibition: The diameter of the clear zones is measured using a ruler or calipers, and the results are recorded.

Comparative Study of Anti-bacterial Activity: Seed Extracts vs. Commercial Anti-bacterial Agents:

Procedure:

- Preparation of Commercial Anti-bacterial Agents: Commercially available anti-bacterial drugs are selected and diluted to obtain different concentrations suitable for testing.
- Preparation of Agar Plates: Agar-agar medium is prepared and poured into sterile Petri dishes, allowed to solidify.
- Inoculation of E. coli Samples: E. coli samples are collected and placed onto the agar plates.
- Testing Procedure: Different concentrations of seed extracts and commercial anti-bacterial agents are applied onto the agar plates with E. coli samples using sterile cotton swabs to ensure uniform application.
- Incubation and Observation: The plates are then incubated at the appropriate temperature for a specified time. After incubation, the plates are observed for any visible effects on bacterial growth, and the zone of inhibition or reduction of bacterial growth is measured.

3. Results and Discussion

Table.1 Phytochemical Analysis of Zea Mays Seeds

Test	Result
Alkaloids (Mayer's Test)	+
Flavonoids (Schinoda Test)	+
Tannins (Ferric Chloride Test)	+
Saponins (Froth Test)	+
Glycosides (Legal's Test)	+
Steroids / Triterpenoids (Liebermann-BurchardTest)	+
Terpenoids (Salkowski Test)	+
Phenols (Ferric Chloride Test)	+

Table.2. In-vitro evaluation of anti-bacterial activity of streptomycin

Micro-organism Inoculum	Extract concentration (µg/ml)	medium	Zone of inhibition(mm)
1 E.coli species	25	Sabouraud Dextrose Agar	2.0 ± 0.3mm
2 E.coli species	50	Sabouraud Dextrose Agar	2.8 ± 0.4mm
3 E.coli species	75	Sabouraud Dextrose Agar	3.9 ± 0.5mm
4 E.coli species	100	Sabouraud Dextrose Agar	4.5 ± 0.4mm

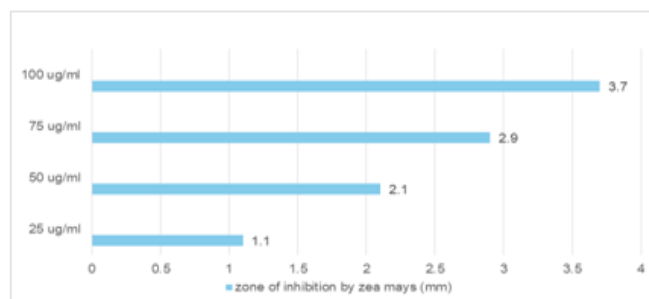


Fig.1. In-vitro evaluation of anti-bacterial activity of Zea mays graph

Micro-organism Inoculum	Extract concentration (µg/ml)	medium	Zone of inhibition(mm)
1 E.coli species	25	Sabouraud Dextrose Agar	2.0 ± 0.3mm
2 E.coli species	50	Sabouraud Dextrose Agar	2.8 ± 0.4mm
3 E.coli species	75	Sabouraud Dextrose Agar	3.9 ± 0.5mm
4 E.coli species	100	Sabouraud Dextrose Agar	4.5 ± 0.4mm

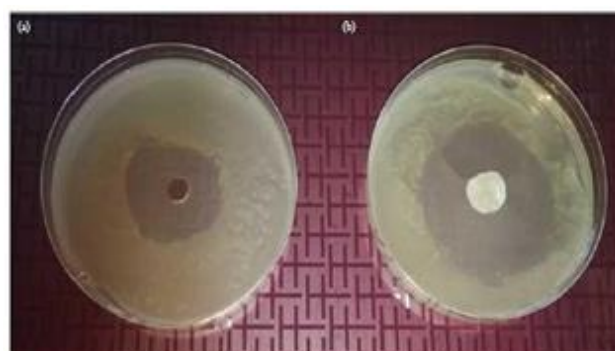


Fig.2. In-vitro evaluation of anti-bacterial activity of streptomycin

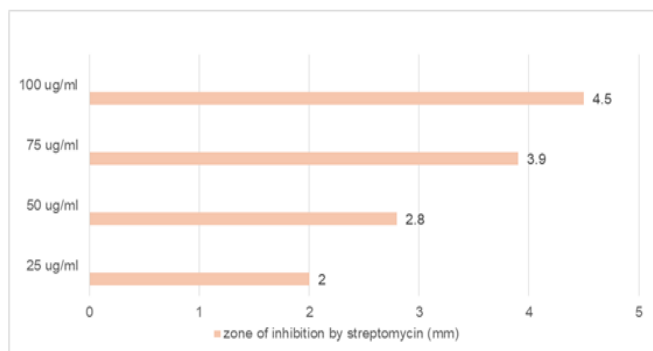


Fig.3. In-vitro evaluation of anti-bacterial activity of streptomycin graph

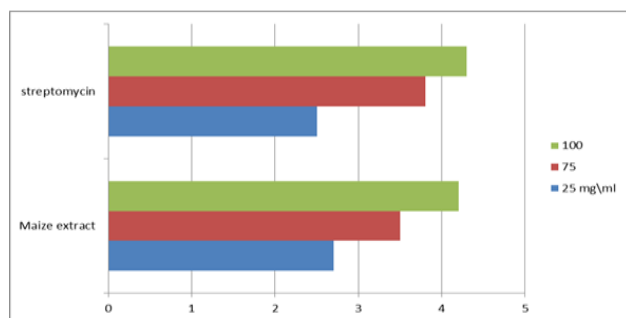


Fig.4. In-vitro evaluation of anti-bacterial activity of streptomycin vs extract Graph

Discussion:

The evaluation of maize extract's anti-bacterial activity represents a significant area of research with implications for both medicine and agriculture. The findings from various studies suggest that maize extract possesses promising anti-bacterial properties, making it a potential natural alternative to conventional antibiotics. This discussion will delve into key findings, explore possible mechanisms of action, and address challenges and future directions in this field. Studies investigating maize extract's anti-bacterial efficacy have revealed varying degrees of effectiveness against different bacterial strains. Notably, maize extracts containing bioactive compounds such as phenols, flavonoids, and alkaloids have demonstrated significant inhibitory effects on both Gram-positive and Gram-negative bacteria. However, the degree of inhibition varies, with Gram-positive bacteria generally being more susceptible to the extract's anti-bacterial activity.

Additionally, the choice of solvent for extraction plays a crucial role, with ethanolic extracts showing greater efficacy compared to aqueous extracts. While the exact mechanisms underlying maize extract's anti-bacterial activity remain unclear, several hypotheses have been proposed. Phenolic compounds may disrupt bacterial cell walls, leading to cell lysis and death, while flavonoids and alkaloids could destabilize bacterial membranes. Another possible mechanism involves the inhibition of bacterial enzymes critical for cell survival, thereby impairing bacterial metabolism and leading to cell death. Further research is needed to elucidate these mechanisms and confirm their role in maize extract's anti-bacterial activity.

Despite its potential, several challenges need to be addressed before maize extract can be widely adopted. Safety and toxicity evaluations are essential to ensure that maize extract does not pose risks to human health or the environment. Additionally, optimizing extraction methods and formulations is crucial for maximizing its anti-bacterial activity.

4. Conclusion

In conclusion, maize extract shows promise as a natural anti-bacterial agent, offering an alternative to conventional antibiotics. Studies have demonstrated its efficacy against various bacterial strains, though further research is needed to fully understand its mechanisms of action and address safety concerns. Future research should focus on identifying specific active compounds, investigating synergistic effects with other agents, and assessing long-term safety and environmental impact. With continued investigation, maize extract could offer valuable contributions to combating bacterial infections and promoting sustainable practices in medicine and agriculture.

5. References

- [1] A.V. Samrot, P. Raji, A.J. Selvarani, P. Nishanthini, Antibacterial activity of some edible fruits and its green synthesized silver nanoparticles against uropathogen – *Pseudomonas aeruginosa* SU 18, *Biocatalysis and Agricultural Biotechnology* 16 (2018) 253–270.
- [2] A.V. Samrot, N. Shobana, R. Jenna, Antibacterial and antioxidant activity of different staged ripened fruit of *Capsicum annum* and its green synthesized silver nanoparticles, *Bionanoscience* 8 (2018) 632–646.
- [3] P. Raji, A.V. Samrot, D.B. Rohan, et al., Extraction, characterization and invitro bioactivity evaluation of alkaloids, flavonoids, saponins and tannins of *Cassia alata*, *Thespesia populnea*, *Euphorbia hirta* and *Wrightia tinctoria*, *Rasayan Journal of Chemistry* 12 (1) (2019) 123–137.
- [4] A.V. Samrot, J.L.A Angalene, S.M. Roshini, S.M., Preethi R, Raji P, Madankumar A, Paulraj P, Kumar SS. Purification, characterization and utilization of polysaccharide of *Araucaria heterophylla* gum for the synthesis of curcumin loaded nanocarrier. *Int. J. Biol. Macromol.* 2019, 140(1): 393-400.
- [5] P. Raji, A.V. Samrot, D. Keerthana, S. Karishma, Antibacterial activity of alkaloids, flavonoids, saponins and tannins mediated green synthesised silver nanoparticles against *Pseudomonas aeruginosa* and *Bacillus subtilis*, *J. Cluster Sci.* (2019).
- [6] K.S. Samanvitha, S.S. Kumar, A.V. Samrot, et al., Targeting acyl Homo serine (AHL) of *Pseudomonas aeruginosa* responsible for biofilm formation using plant metabolites, *J. Pure Appl. Microbiol.* 13 (3) (2019).
- [7] P. Senthilkumar, S. Rashmitha, P. Veera, C.V. Ignatious, C. SaiPriya, A.V. Samrot. Antibacterial

- activity of neem extract and its green synthesized silver nanoparticles against *Pseudomonas aeruginosa*. *J. Pure Appl. Microbiol.* 12(02).2018.
- [8] A.V. Samrot, Silky, C.V. Ignatious, P. Raji, C. SaiPriya, J.A. Selvarani, Bioactivity studies of datura metel, aegle marmelos, annona reticulata and saraca indica and their green synthesized silver nanoparticle, *J. Pure Appl. Microbiol.* 13 (1) (2019).
- [9] J. Yang, X. Li, Y. Xue, N. Wang, W. Liu, Anti-hepatoma activity and mechanism of corn silk polysaccharides in H22 tumor-bearing mice, *Int. J. Biol. Macromol.* 64 (2014) 276–280.
- [10] Z. Maksimovic, D. Malencic N. Kovacevic, Polyphenol contents and antioxidant activity of *Maydis stigma* extracts, *Bioresour. Technol.* 96 (2005) 873–877.
- [11] F. Nessa, Z. Ismail, N. Mohamed, Antimicrobial activities of extracts and flavonoid glycosides of corn silk (*Zea mays L.*), *Int. J. Biotechnol. Wellness Ind.* 1 (2) (2012) 115–120.
- [12] D.V.O. Velazquez, H.S. Xavier, J.E.M. Batista, C. de CastroChavas, *Zea mays L.* extracts modify glomerular function and potassium urinary excretion in conscious rats, *Phytomedicine* 12 (2005) 363–369.
- [13] A. El-Ghorab, K.F. El-Massry, K. Shibamoto, Chemical composition of the volatile extract and antioxidant activities of the volatile and nonvolatile extracts of Egyptian corn silk (*Zea mays L.*), *J. Agric. Food Chem.* 55 (2007) 9124–9127.
- [14] T. Namba, H. Xu, S. Kadota, M. Hattori, T. Takahashi, Y. Kojima, Inhibition of Ig E formation in mice by glycoproteins from corn silk, *Phytother Res.* 7 (1993): 227–230.
- [15] M.P. Johnson, A.V. Ruban, *Arabidopsis* plants lacking PsbS protein possess photoprotective energy dissipation, *Plant J.* 61 (2) (2010) 283–289.
- [16] H.F.S. Akrayi, Z.F.A. Abdulrahman, Evaluation of the antibacterial efficacy and the phytochemical analysis of some plant extracts against human pathogenic bacteria, *JPCS* (7) (2013): 29–39.