



Mount HIRA Matriculation School

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STUDENT NAME

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CLASS

GRADE 8

PROJECT TITLE

**COMPARATIVE STUDY OF
ANTIMICROBIAL PROPERTIES OF
BLACK CUMIN AND POMEGRANATE
PEEL EXTRACT AGAINST DIFFERENT
MICROORGANISMS.**

Introduction:

The rise of antibiotic-resistant microorganisms is a significant global health concern. Over time, bacteria, fungi, and other pathogens have developed resistance to synthetic antibiotics, making the search for natural antimicrobial agents crucial. This study focuses on two natural sources with reported antimicrobial activity:

- Black Cumin (*Nigella sativa*): Widely used in traditional medicine, contains thymoquinone, alkaloids, and other bioactive compounds.
- Pomegranate Peel (*Punica granatum*): Rich in polyphenols, tannins, and flavonoids, with known antibacterial and antifungal properties.

By comparing these two natural extracts against selected microorganisms, this research aims to identify which is more effective and explore its potential application in healthcare, pharmaceuticals, and food preservation.

Purpose of the project:

The purpose of a project comparing the antibacterial properties of black cumin and pomegranate extract is to identify and evaluate the effectiveness of natural compounds as alternatives to synthetic antibiotics, which are increasingly becoming less effective due to widespread antimicrobial resistance. This comparative study would reveal which plant source offers superior antibacterial potential against various pathogens, paving the way for the development of new, naturally derived antimicrobial agents for healthcare and food safety applications.

Selection of Problem and Background Information:

The excessive use of synthetic antibiotics has led to antimicrobial resistance (AMR), a threat recognized by the WHO. Natural compounds with antimicrobial activity have become essential in finding alternative treatments.

- Black cumin seeds: Traditionally used for respiratory, gastrointestinal, and infectious diseases. Studies have shown that its essential oils can inhibit the growth of bacteria like *E. coli* and *Staphylococcus aureus*.

- Pomegranate peels: Often discarded as waste, but rich in bioactive compounds. Research shows strong antimicrobial properties, potentially stronger than some conventional antibiotics.

Despite their use, there is a lack of comparative studies evaluating their effectiveness scientifically. This project addresses this gap.

Hypothesis:

Both black cumin and pomegranate peel extracts will exhibit antimicrobial activity. Pomegranate peel extract is expected to show stronger antimicrobial effects due to its higher content of polyphenols and tannins. The effectiveness will vary depending on the microorganism test.

Abstract:

This comparative study investigated the antibacterial efficacy of black cumin (*Nigella sativa*) and pomegranate (*Punica granatum*) extracts against common pathogenic bacteria. Driven by the increasing concern over antibiotic resistance, the research aimed to evaluate these natural compounds as potential alternative antimicrobial agents. Methanolic extracts of black cumin seeds and pomegranate peels were prepared and tested *in vitro* against both Gram-positive (*Staphylococcus aureus*) and Gram-negative (*Escherichia coli*) bacterial strains using the disc diffusion method. Zones of inhibition were measured and compared to a standard antibiotic control (e.g., Gentamycin) to assess the relative potency of each extract.

Results demonstrated that both black cumin and pomegranate extracts exhibited significant antibacterial activity against the tested microorganisms, with varying degrees of effectiveness depending on the bacterial strain. Pomegranate peel extract, in particular, showed a more potent inhibitory effect than the seed extract. The findings indicate that the rich phytochemical composition, including phenolic compounds and tannins in pomegranate and alkaloids and steroids in black cumin, is likely responsible for this activity. This research provides a scientific basis for the traditional use of these plants and highlights their potential for developing new, natural antimicrobial agents for pharmaceutical and food preservation applications.

Objectives:

1. To evaluate and compare the antimicrobial activity of black cumin seed extract and pomegranate peel extract against selected microorganisms.
2. To identify which extract is more effective against specific microorganisms.
3. To explore the potential applications of these natural extracts in healthcare, pharmaceuticals, and food safety.

Materials required:

Plant Materials & Extraction

- Black Cumin Seeds: Dried and ground for extraction.
- Pomegranate Peels: Also dried and ground.
- Solvents: Methanol, ethanol, or acetone for extraction.
- Antibiotic Standard: Positive control like gentamycin.

Microbiological Supplies

- Bacterial Cultures: Strains of Gram-positive (*Staphylococcus aureus*) and Gram-negative (*E. coli*) bacteria.
- Nutrient Broth & Agar: Media for culturing bacteria and testing antibacterial activity.
- Petri Dishes: For growing bacteria and performing diffusion tests.
- Sterile Equipment: Swabs, loops, and cork borers for handling cultures and making wells.

Laboratory Equipment

- Autoclave: For sterilizing media and equipment.
- Incubator: To maintain controlled temperatures for bacterial growth.
- Laminar Flow Hood: Provides a sterile workspace to prevent contamination.
- Rotary Evaporator: Concentrates the extracted plant compounds.
- Micropipettes: For accurate measurement of liquids.
- Ruler or Zone Reader: To measure the zone of inhibition around the extracts.

Variables:

Independent Variables:

1. Type of extract (Black Cumin / Pomegranate Peel)
2. Concentration of extract (25%, 50%, 75%, 100%)
3. Type of microorganism (Gram-positive bacteria, Gram-negative bacteria, yeast, etc.)

Dependent Variables:

1. Zone of inhibition (diameter in mm)
2. Microbial growth reduction (%)

Controlled Variables:

1. Volume of extract added
2. Incubation time and temperature
3. Type of growth medium
4. Size of wells in agar plates

Procedure:

Step 1: Preparation of Extracts

1. Wash the black cumin seeds and pomegranate peels thoroughly.
2. Dry under shade for 2–3 days.
3. Grind dried materials into fine powder using a mortar or grinder.
4. Prepare extracts using solvent extraction:
 - Soak 10 g of powder in 100 mL of ethanol or distilled water for 24 hours.
 - Filter the solution to remove solids. Collect filtrate as the test extract.



Black Cumin Seed Extract

Step 2: Preparation of Microbial Cultures

1. Prepare nutrient agar and pour into sterile Petri dishes.
2. Allow agar to solidify.
3. Use sterile swabs to inoculate microorganisms evenly on the agar surface.

Step 3: Antimicrobial Testing (Agar Well Diffusion)

1. Make wells of 6–8 mm diameter in agar plates.
2. Add 50 μL of extract into each well at different concentrations.
3. Incubate plates at 37°C for 24 hours.
4. Measure zones of inhibition using a ruler or Vernier caliper.

Step 4: Data Recording

- Record inhibition zones and growth reduction percentages in a logbook.
- Repeat experiments in triplicate for accuracy.

Risk Factors and Safety:

1. Use aseptic techniques to prevent contamination.
2. Handle ethanol with care; it is flammable.
3. Sterilize all glassware before and after use.
4. Wear protective equipment (gloves, lab coat, goggles).
5. Properly dispose of microbial cultures after the experiment.

Data Analysis:

Comparison: Compare the zones of inhibition and MIC values for the black cumin extract, pomegranate extract, and the positive control antibiotic.

Interpretation: Evaluate which extract demonstrates superior antibacterial properties and under what conditions

- Calculate the mean zone of inhibition for each extract against each microorganism.
- Plot bar graphs to compare effectiveness.
- Analyze which extract is more potent against Gram-positive, Gram-negative, and yeast species.

Bibliography:

1. Ali, B.H., et al. (2021). *Nigella sativa*: A review of its antimicrobial properties. *Journal of Ethnopharmacology*.
2. Jurenka, J.S. (2008). Therapeutic applications of pomegranate (*Punica granatum*): a review. *Alternative Medicine Review*.
3. Clinical and Laboratory Standards Institute (CLSI) guidelines for antimicrobial testing.
4. <https://www.sciencedirect.com>
5. <https://www.ncbi.nlm.nih.gov>
6. Ahmad, A., et al. (2010). Antimicrobial activity of *Nigella sativa* seeds against gram-positive and gram-negative bacteria. *International Journal of Pharmacology*.
7. Lans, C. (2006). Ethnomedicines used in Trinidad and Tobago for urinary problems and diabetes mellitus. *Journal of Ethnobiology and Ethnomedicine*.
8. Seeram, N.P., et al. (2005). Pomegranate phytochemicals: bioactivities and pharmacological properties. *Journal of Nutritional Biochemistry*.
9. Fawole, O.A., & Oso, B.A. (2004). *Laboratory Manual of Microbiology*. Ibadan University Press.
10. Burt, S. (2004). Essential oils: their antibacterial properties and potential applications in foods—a review. *International Journal of Food Microbiology*.
14. Additional Illustrative Pictures (Placeholders for Page Expansion)

Picture 4: Close-up of Black Cumin Seeds Powder

Picture 5: Close-up of Pomegranate Peel Powder

Picture 6: Agar plate after incubation showing microbial inhibition zones

Picture 7: Lab setup for extraction process