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Research Article

Design and Evaluation of Herbal Anti-Tick Effervescent Bath Bomb containing *Azadirachta indica* and *Allium sativum* Extract

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ABSTRACT

The present study focuses on the development and validation of a simple, precise, and reliable analytical method for the estimation of brivaracetam in bulk and tablet dosage form using reverse phase high performance liquid chromatography (RP-HPLC). The primary objective of this work was to establish an efficient chromatographic method suitable for routine quality control analysis. Chromatographic separation was achieved using a c18 column with a mobile phase consisting of methanol, acetonitrile, and sulphate buffer in the ratio of 15:35:50. The analysis was carried out at a detection wavelength of 208 nm. The optimized conditions provided a well-defined chromatogram with good peak shape and resolution for brivaracetam. The developed method was validated according to ICH guidelines, demonstrating acceptable levels of accuracy, precision, linearity, and specificity. The method showed consistent and reproducible results for the quantification of brivaracetam in both bulk and tablet formulations.

INTRODUCTION

Ticks are blood-sucking ectoparasites that infest animals, such as dogs, cattle, and other domestic pets. They are responsible for skin irritation, allergic reactions, anemia, and transmission of serious diseases such as tick fever, Lyme disease, and babesiosis. Tick infestation not only affects animal health but also reduces their productivity and overall well-being. Therefore, effective tick

control is essential for veterinary care and hygiene management. [1]

Conventionally, tick control is achieved using synthetic chemical agents, such as shampoos, sprays, powders, and spot-on treatments containing insecticides. Although these products are effective, their prolonged use may lead to several disadvantages, including skin irritation, toxicity, development of resistance in ticks, and

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environmental pollution. In addition, some chemical agents may leave harmful residues on animal skin and fur, posing risks to both animals and humans.[2]



Fig 1.1 :- Ticks presents on animals are given above.

In recent years, there has been a growing interest in the use of herbal and natural products due to their safety, biodegradability, and minimal side effects. Herbal ingredients such as neem oil, eucalyptus oil, and citronella oil are widely recognized for their insecticidal, antimicrobial, and repellent properties. Neem oil contains active constituents like azadirachtin, which interfere with the growth and reproduction of ticks. Eucalyptus oil acts as a natural repellent and provides a cooling effect, while citronella oil is well known for its ability to repel insects effectively.

A bath bomb is a solid, compact formulation that typically consists of sodium bicarbonate and citric acid. When added to water, it undergoes an effervescent reaction, releasing carbon dioxide gas and producing a fizzing effect. This effervescence helps in the uniform dispersion of active ingredients in water, enhancing their contact with the animal's skin and fur. Bath bombs are widely used in cosmetic and personal care products due to their ease of use, aesthetic appeal, and therapeutic benefits.

The concept of incorporating herbal anti-tick agents into a bath bomb formulation offers a novel and convenient approach for tick control. The effervescent action ensures better distribution of herbal oils, thereby improving their efficacy.

Additionally, the use of natural ingredients makes the formulation safer for regular use without causing harm to the animal or the environment.[3]

The formulation of a herbal anti-tick bath bomb involves careful selection of ingredients to ensure stability, effectiveness, and user acceptability. Along with active herbal components, excipients such as binders, fillers, and stabilizers are used to maintain the structural integrity and performance of the bath bomb. Evaluation of the prepared formulation is essential to assess parameters such as physical appearance, hardness, pH, effervescence time, moisture content, stability, and anti-tick activity.

Thus, the development of a herbal anti-tick bath bomb represents an innovative, eco-friendly, and user-friendly alternative to conventional tick control methods. It combines the benefits of herbal medicine with modern formulation techniques to provide a safe and effective solution for managing tick infestations in animals.[4]

1.1 Introduction to Herbal Veterinary Care

The use of herbal medicines for the treatment and prevention of diseases has been practiced since ancient times. Medicinal plants have played an important role in maintaining the health of both humans and animals. In recent years, there has

been increasing interest in herbal veterinary formulations because of their safety, eco-friendliness, cost effectiveness, and reduced side effects compared to synthetic chemical products. Herbal products are widely used in the management of parasitic infestations, skin infections, wounds, and insect bites in domestic animals and pets.

Herbal formulations containing plant extracts with insecticidal and antimicrobial properties are considered promising substitutes for synthetic anti-tick agents. Among these, extracts obtained from *Azadirachta indica* (Neem) and *Allium sativum* (Garlic) have gained significant attention because of their potent antiparasitic, antimicrobial, antifungal, and insect-repellent activities.

The present project focuses on the formulation and evaluation of a herbal anti-tick bath bomb using neem and garlic extracts. Bath bombs are solid effervescent formulations that dissolve in water and release active ingredients uniformly. Such formulations can provide convenient, effective, and pleasant application during animal bathing while ensuring better distribution of herbal extracts over the skin and fur.

1.2. Ticks and Their Importance

Ticks are blood-sucking ectoparasites belonging to the class Arachnida and order Acari. They infest a wide range of animals including dogs, cattle, buffaloes, sheep, goats, horses, and poultry. Ticks attach themselves to the skin of animals and feed on blood for prolonged periods. During feeding, they can transmit pathogens responsible for various diseases.

Tick infestation is one of the major veterinary problems worldwide. Warm and humid environmental conditions favor the rapid growth and reproduction of ticks. In tropical countries like

India, tick infestation is highly prevalent and causes major economic losses in livestock production.

Harmful Effects of Tick Infestation

1. Blood loss leading to anemia
2. Skin irritation and itching
3. Hair loss and wounds
4. Reduced milk and meat production
5. Weakness and stress in animals
6. Transmission of tick-borne diseases
7. Secondary bacterial infections

Ticks are vectors for many serious diseases such as:

- Babesiosis
- Theileriosis
- Ehrlichiosis
- Lyme disease
- Rocky Mountain spotted fever

Because of these harmful effects, effective tick control measures are essential for maintaining animal health and productivity.

1.3. Conventional Anti-Tick Treatments and Their Limitations

Several synthetic chemical acaricides are available in the market for tick control. These include:

- Organophosphates
- Carbamates
- Pyrethroids
- Amitraz
- Ivermectin preparations

Although these products are effective initially, prolonged use has resulted in many disadvantages.

Limitations of Synthetic Anti-Tick Products

Development of Resistance

Ticks develop resistance after repeated exposure to chemical acaricides, reducing treatment effectiveness.

Toxicity

Synthetic chemicals may cause toxicity in animals, especially when used improperly or in high concentrations.

Skin Irritation

Many chemical products produce dryness, itching, redness, and irritation on animal skin.

Environmental Pollution

Chemical residues contaminate soil and water bodies and may affect non-target organisms.

Residual Effects

Chemical residues may remain in milk, meat, and animal products, creating public health concerns.

These disadvantages have encouraged researchers to explore herbal alternatives for safer tick management.

1.4. Herbal Medicines as Alternatives

Herbal medicines are preparations obtained from medicinal plants containing biologically active compounds. Plant-based products are considered safer because they are biodegradable and generally produce fewer adverse effects.

Herbal formulations are increasingly preferred because:

- They are naturally available
- Economical and easy to prepare
- Environment friendly
- Less toxic to animals
- Less likely to produce resistance
- Possess multiple pharmacological activities

Medicinal plants contain secondary metabolites such as:

- Alkaloids
- Flavonoids
- Tannins
- Terpenoids
- Saponins
- Phenolic compounds

These phytochemicals exhibit insecticidal, antimicrobial, antiparasitic, and antioxidant activities.

Among the various medicinal plants, neem and garlic are widely recognized for their anti-tick and insect-repellent properties.



1.5. *Azadirachta indica* (Neem)

Azadirachta indica is commonly known as Neem and belongs to the family Meliaceae. Neem is considered one of the most important medicinal plants in India and has been used in Ayurveda, Siddha, and Unani systems of medicine for centuries.

Almost every part of the neem tree including leaves, bark, seeds, flowers, and oil possesses medicinal value.

Chemical Constituents of Neem

Neem contains several active compounds such as:

- Azadirachtin
- Nimbin
- Nimbidin
- Salannin
- Gedunin
- Quercetin

Among these, azadirachtin is mainly responsible for insecticidal and repellent activity.

Pharmacological Activities of Neem

Neem possesses:

- Antibacterial activity
- Antifungal activity
- Antiviral activity
- Anti-inflammatory activity
- Insecticidal activity
- Antiparasitic activity

Anti-Tick Properties of Neem

Neem acts against ticks by:

- Repelling ticks
- Inhibiting feeding
- Disturbing growth and reproduction
- Causing mortality in immature stages

Neem-based formulations are safer for animals and the environment compared to synthetic acaricides.

1.6. *Allium sativum* (Garlic)

Allium sativum belongs to the family Amaryllidaceae and is widely used as both food and medicine. Garlic has been used traditionally for treating infections, cardiovascular disorders, and parasitic infestations.

Garlic contains sulfur-containing compounds responsible for its characteristic odor and medicinal properties.

Chemical Constituents of Garlic

Major active constituents include:

- Allicin
- Alliin
- Ajoene
- Diallyl sulfide
- Selenium compounds

Pharmacological Activities of Garlic

- Garlic exhibits:
- Antibacterial activity



- Antifungal activity
- Antiviral activity
- Antioxidant activity
- Antiparasitic activity
- Insect-repellent activity

Anti-Tick Activity of Garlic

Garlic extract acts by:

- Repelling ticks and insects
- Interfering with parasite metabolism
- Producing toxic effects on ectoparasites

Garlic also helps reduce microbial infections associated with tick bites.

1.7.Synergistic Effect of Neem and Garlic

Combining neem and garlic extracts may produce enhanced anti-tick activity because both plants contain bioactive compounds with complementary mechanisms of action.

Advantages of Combination Therapy

1. Broader antiparasitic activity
2. Improved repellency
3. Reduced chances of resistance
4. Enhanced antimicrobial protection
5. Better skin health maintenance

The combination may therefore provide effective and natural tick control in animals.

1.8.Bath Bomb Drug Delivery System

Bath bombs are solid effervescent formulations composed mainly of acids and bicarbonates that react in water to release carbon dioxide gas. This effervescence helps disperse active ingredients uniformly in bathing water.

Composition of Bath Bombs

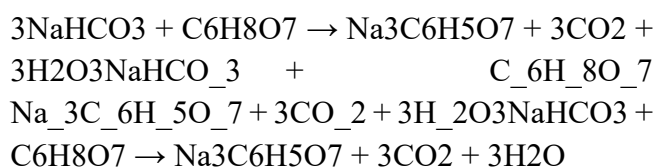
Common ingredients include:

- Citric acid
- Sodium bicarbonate
- Essential oils
- Herbal extracts
- Starch or binders
- Colourings and fragrance agents

Mechanism of Effervescence

When the bath bomb comes in contact with water, citric acid reacts with sodium bicarbonate to produce carbon dioxide bubbles.

The reaction can be represented as:



This effervescence:

- Enhances mixing of herbal extracts
- Improves contact with animal skin and fur
- Provides easy and convenient application

1.9.Advantages of Herbal Anti-Tick Bath Bombs

The herbal anti-tick bath bomb offers several benefits:

Easy Application

The formulation can be easily dissolved in water during animal bathing.

Uniform Distribution

Effervescence helps distribute active ingredients evenly.

Eco-Friendly

Herbal ingredients are biodegradable and environmentally safe.

Reduced Toxicity

The formulation is safer than synthetic chemicals.

Pleasant Bathing Experience

Essential oils and herbal ingredients improve odor and cleanliness.

Improved Animal Hygiene

The formulation may also reduce microbial load and skin infections.

1.10. Need for the Present Study

Tick infestation remains a major problem in veterinary practice despite the availability of several synthetic acaricides. Resistance development, toxicity, environmental hazards, and high costs associated with chemical products highlight the need for alternative herbal formulations.

Neem and garlic are well-known medicinal plants with proven antiparasitic activities. However, limited research has been conducted on combining

these extracts in an effervescent bath bomb dosage form for veterinary use.

The present study aims to formulate and evaluate a herbal anti-tick bath bomb containing neem and garlic extracts to provide:

- Effective tick control
- Safer animal application
- Eco-friendly treatment
- Cost-effective herbal alternative

This formulation may help improve animal health and hygiene while minimizing the harmful effects associated with synthetic anti-tick products.

1.11. Scope of the Study

The study includes:

- Extraction of neem and garlic bioactive constituents
- Formulation of herbal bath bombs
- Evaluation of physicochemical properties
- Assessment of anti-tick activity
- Stability and safety studies

The project may contribute to the development of herbal veterinary formulations and encourage the use of natural products in parasite control.

2. REVIEW OF LITERATURE

1. Biswas K., Chattopadhyay I., Banerjee R. K., and Bandyopadhyay U. et.al., (2002) extensively reviewed the biological activities of neem (*Azadirachta indica*). Their study reported that neem contains active constituents such as azadirachtin, nimbin, and nimbidin, which possess



insecticidal, antimicrobial, antifungal, and anti-inflammatory properties. The researchers concluded that neem interferes with insect growth and reproduction and acts as an effective natural acaricide against ticks and parasites.

2. Ankri S. and Mirelman D. et.al., (1999) studied the antimicrobial properties of garlic (*Allium sativum*). Their research showed that garlic contains sulfur compounds such as allicin, ajoene, and diallyl sulphides, which exhibit antibacterial, antifungal, antiparasitic, and insect-repellent activities. They concluded that garlic extract can serve as a natural protective agent against microorganisms and parasites.

3. Isman M. B. et.al., (2006) reviewed botanical insecticides and repellents in modern agriculture and veterinary applications. The study highlighted that plant-derived products such as neem oil, citronella oil, and garlic extract are effective alternatives to synthetic pesticides. According to the researcher, herbal products are biodegradable, eco-friendly, and safer for animals and humans.

4. Nerio L. S., Olivero-Verbel J., and Stashenko E. et.al., (2010) investigated the repellent activity of essential oils. Their review reported that essential oils obtained from plants exhibit strong insect-repellent properties because of their volatile aromatic compounds. The study emphasized that herbal oils can effectively repel ticks and insects without causing severe toxicity.

5. Pavela R. et.al., (2015) studied the role of essential oils in eco-friendly pest management. The research demonstrated that plant-derived oils possess insecticidal, larvicidal, antimicrobial, and antioxidant activities. The author concluded that herbal formulations can reduce dependence on synthetic chemicals and help protect the environment.

6. George J. E., Pound J. M., and Davey R. B. et.al., (2004) discussed chemical control of ticks and resistance development. Their study reported that repeated use of synthetic acaricides has resulted in resistant tick populations, reducing effectiveness of conventional products. The researchers recommended development of alternative tick-control methods using herbal agents.

7. Ghosh S., Tiwari S. S., Sharma A. K., and coworkers et.al., (2015) reviewed tick distribution and control strategies in India. Their work emphasized the increasing importance of herbal formulations and integrated pest management techniques because of growing resistance and toxicity associated with chemical acaricides.

8. Bakkali F., Averbeck S., Averbeck D., and Idaomar M. et.al., (2008) studied the biological activities of essential oils. They reported that essential oils possess antimicrobial, antioxidant, antifungal, and insecticidal properties. Their study also suggested that essential oils can penetrate insect membranes and affect nervous system activity, resulting in repellency and mortality.

9. Aulton M. E. and Taylor K. et.al., (2017) explained the pharmaceutical importance of effervescent dosage forms. According to their research, sodium bicarbonate and citric acid react in water to release carbon dioxide, improving dispersion and delivery of active ingredients. Effervescent systems also enhance user compliance and provide rapid action.

10. Rowe R. C., Sheskey P. J., and Quinn M. E. et.al., (2009) described the role of pharmaceutical excipients in formulation development. Their work highlighted that sodium bicarbonate, citric acid, starch, and magnesium sulphate are important excipients in effervescent preparations



because they improve stability, hardness, dispersion, and functionality.

3. NEED OF WORK

Ticks are common ectoparasites that affect animals like dogs, cattle, and pets, causing itching, skin irritation, inflammation, and diseases such as Lyme disease and babesiosis. Chemical anti-tick products like sprays and shampoos are effective but may cause toxicity, skin allergies, environmental pollution, and resistance in ticks after prolonged use.[5]

Herbal ingredients such as neem oil, eucalyptus oil, and citronella oil are safer and eco-friendly alternatives because of their insecticidal and repellent properties. However, herbal oils and sprays may have drawbacks like difficult application and uneven distribution.[6]

To overcome these problems, a herbal anti-tick bath bomb was developed as an innovative formulation. Bath bombs dissolve in water and release herbal ingredients uniformly through effervescence, improving contact with the animal's skin and fur. They are easy to use, safe, economical, and environmentally friendly.

This study aims to formulate and evaluate a herbal anti-tick bath bomb as a convenient and effective alternative to conventional chemical-based anti-tick products.[7]

4. AIMS AND OBJECTIVES

Aim:

The primary aim of the present work is to formulate and evaluate a herbal anti-tick bath bomb using natural ingredients that is safe, effective, eco-friendly, and convenient for controlling tick infestation in animals.

Objectives

To achieve the above aim, the following specific objectives are undertaken:

1. Selection of Herbal Ingredients: -

- To select suitable herbal ingredients such as neem oil, eucalyptus oil, and citronella oil.
- To utilize their insecticidal, repellent, and antimicrobial properties for tick [8]
- Control.

2. Formulation Development: -

- To develop an effervescent bath bomb using sodium bicarbonate and citric acid.
- To incorporate herbal oils uniformly into the formulation.
- To obtain a stable and aesthetically acceptable product.

3. Optimization of Formulation: -

- To adjust proportions of ingredients for proper hardness and shape.
- To ensure controlled effervescence and complete dissolution.
- To improve fragrance and user acceptability.[9]

4. Evaluation of Physical Properties: -

- To evaluate organoleptic characteristics such as colour, odour, and appearance.
- To determine hardness, friability, and weight variation.



- To assess moisture content and stability.

5. Evaluation of Chemical Properties: -

- To determine the pH of the formulation.
- To ensure compatibility with animal skin.
- To check for any chemical instability.[10]

6. Performance Evaluation: -

- To study effervescence time and dispersion behaviour in water.
- To evaluate foamability and solubility.

7. Anti-Tick Efficacy Study: -

- To assess the effectiveness of the formulation against ticks.
- To evaluate repellent and acaricidal activity of herbal ingredients.

8. Safety Evaluation: -

- To perform skin irritation test on animals.
- To ensure the formulation is non-toxic and safe for regular use.

9. Stability Studies: -

- To evaluate the formulation under different environmental conditions.
- To study changes in physical appearance, fragrance, and effectiveness over.
- Time.

10. Development of Eco-Friendly Product: -

- To create a biodegradable and environmentally safe formulation.

- To reduce dependency on synthetic chemical products.[11]

5. PLAN OF WORK

The present research work on the formulation and evaluation of a herbal anti-tick bath bomb is systematically planned to ensure scientific accuracy, reproducibility, and effectiveness of the final product. The work is divided into several well-defined stages, starting from literature review to final evaluation and documentation.

1. Literature Survey

A comprehensive literature review is carried out to understand:

- The biology and life cycle of ticks.
- Harmful effects of tick infestation on animals.
- Conventional anti-tick treatments and their limitations.
- Herbal alternatives with anti-tick properties

Scientific journals, textbooks, and research articles are referred to study the efficacy of herbal ingredients such as neem oil, eucalyptus oil, and citronella oil. Additionally, previous studies on effervescent formulations like bath bombs are reviewed to understand formulation techniques.

2. Selection and Procurement of Materials

Based on the literature review, suitable materials are selected:

Active Ingredients: -

- Neem oil – insecticidal and acaricidal
- Eucalyptus oil – repellent and antimicrobial



- Citronella oil – insect repellent

Excipients: -

- Sodium bicarbonate – effervescence base
- Citric acid – acid component
- Corn starch – binder and stabilizer
- Epsom salt – improves texture

All materials are procured from reliable sources and checked for quality and purity.

3. Preformulation Studies

Preformulation studies are performed to ensure compatibility and stability of ingredients:

- Physical properties (colour, odour, texture).
- Solubility studies.
- Compatibility between herbal oils and excipients.
- Determination of suitable ratios.

These studies help in designing a stable and effective formulation.

4. Formulation Development and Optimization

Different trial formulations are prepared by varying proportions of ingredients to achieve:

- Proper hardness and shape.
- Controlled effervescence.
- Uniform distribution of herbal oils.

Optimization is done to obtain the best formulation with desired properties such as stability, appearance, and performance.

5. Method of Preparation

The bath bomb is prepared using the following steps:

- Dry ingredients (sodium bicarbonate, citric acid, corn starch, Epsom salt) are accurately weighed.
- Ingredients are mixed thoroughly to ensure uniformity.
- Herbal oils are added gradually with continuous mixing.
- A small quantity of water or binding agent is added to achieve proper consistency.
- The mixture is pressed into moulds to obtain desired shape.
- The moulded bath bombs are dried for 24–48 hours.
- Dried bath bombs are removed and stored in airtight containers.

6. Evaluation of Formulation

The prepared bath bombs are evaluated using various parameters:

The prepared bath bombs are evaluated using various parameters:

A. Organoleptic Evaluation

1. Colour
2. Odour
3. Appearance

B. Physical Evaluation

1. Hardness and friability

2. Weight variation

3. Shape and size

C. Chemical Evaluation

1. pH determination

D. Performance Evaluation

1. Effervescence time

2. Solubility and dispersion

3. Foamability

7. Anti-Tick Efficacy Study

The effectiveness of the formulation is evaluated by:

1. Observing tick repellent activity.

2. Studying tick mortality rate.

3. Comparing results with standard treatments.

4. This step confirms the functional performance of the herbal bath bomb.

8. Safety and Skin Irritation Study

1. Patch test is performed on animal skin.

2. Observations are made for redness, irritation, or allergic reactions.

3. Ensures safety for regular use.

9. Stability Studies

Stability testing is carried out under different environmental conditions:

1. Room temperature

2. High temperature

3. Humidity conditions

4. Parameters observed:

5. Change in colour

6. Loss of fragrance

7. Reduction in efficacy

10. Data Recording and Analysis

1. All observations are recorded systematically.

2. Data is analysed and compared with standard values.

3. Graphs and tables may be used for better presentation.

11. Interpretation of Results

1. Results are critically analysed.

2. Effectiveness of formulation is evaluated.

3. Advantages over conventional products are discussed.

12. Documentation and Report Preparation

1. All experimental work is compiled.

2. Results are presented in structured format.

3. Black book is prepared with proper headings.

4. Final review and corrections are done.

6. DRUG AND EXCIPIENT PROFILE

1. Azadirachta Indica





Fig 6.1. Neem (Azadirachta Indica)

Biological Source: -

Obtained from seeds of Azadirachta indica.

Family: -

Meliaceae

Chemical Constituents: -

1. Azadirachtin
2. Nimbin

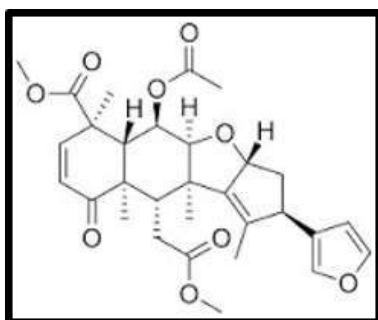


Fig 6.2 Structure of Nimbin

3. Nimbidin



Fig 6.3 Structure of Nimbidin

Properties

1. Insecticidal
2. Acaricidal
3. Antimicrobial
4. Anti-inflammatory

Mechanism of Action

Azadirachtin interferes with the growth and reproduction of ticks, inhibiting feeding and progression of lifecycle.

Uses

1. Anti-tick agent
2. Skin protection
3. Antiseptic

Advantages

1. Natural and biodegradable
2. Safe for animals
3. Minimal side effects

2. Allium Sativum



Fig 6.4 Garlic (Allium Sativum)

Biological Source

Allium sativum consists of the fresh or dried bulbs of garlic obtained from the plant Allium sativum Linn.

Family

It belongs to the family Amaryllidaceae (previously classified under Liliaceae).

Chemical Constituents

Major chemical constituents of garlic are:

1. Alliin
2. Allicin (main active constituent)
3. Sulfur-containing compounds
4. Diallyl sulphide
5. Diallyl disulphide
6. Ajoene
7. Volatile oils
8. Proteins
9. Enzymes
10. Vitamins and minerals

Properties

1. Antibacterial
2. Antifungal
3. Antiviral
4. Antioxidant
5. Anti-inflammatory
6. Antihypertensive

7. Hypolipidemic (reduces cholesterol)

Mechanism of Action

When garlic cloves are crushed, the enzyme alliinase converts alliin into allicin.

3. Allicin:

1. Inhibits growth of microorganisms.
2. Reduces cholesterol synthesis.
3. Prevents platelet aggregation.
4. Acts as an antioxidant by scavenging free radicals.

Uses

1. Used in treatment of hypertension.
2. Helps reduce cholesterol level.
3. Prevents cardiovascular diseases.
4. Used as antimicrobial agent.
5. Improves immunity.
6. Used in cough and cold preparations.

Advantages

1. Natural and easily available remedy.
2. Broad spectrum antimicrobial action.
3. Fewer side effects compared to synthetic drugs.
4. Economical.
5. Provides cardiovascular protection.
6. Possesses antioxidant benefits.



II. EXCIPIENT PROFILE

1. Sodium Bicarbonate

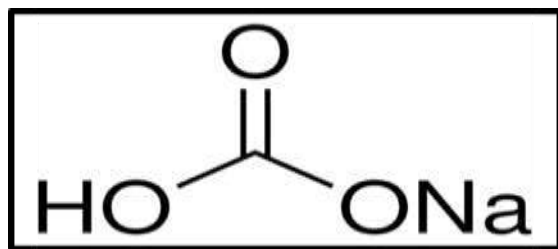


Fig 6.5 Structure of Sodium Bicarbonate

Category

Effervescent agent (base)

Properties

1. White crystalline powder. 2. Water-soluble.

Function

1. Reacts with citric acid to produce carbon dioxide (effervescence).
2. Role in Formulation.
3. Provides fizzing action.
4. Helps in dispersion of active ingredients.

2. Citric Acid

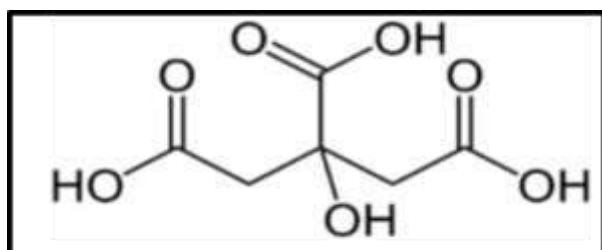


Fig 6.6 Structure of Citric Acid

Category

Acidulant

Properties

1. Colorless crystalline solid
2. Sour taste

Function

Reacts with sodium bicarbonate in presence of water.

Role

1. Produces effervescence.
2. Maintains PH.

3. Corn Starch

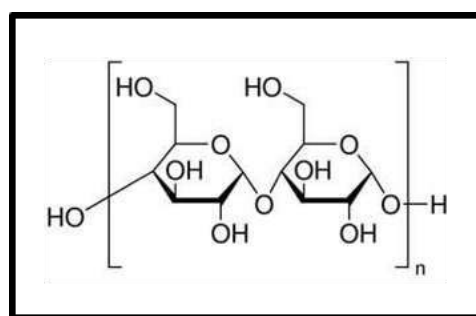


Fig 6.7. Structure Of Corn Starch

Category

Binder / Filler

Properties

1. Fine white powder
2. Absorbs moisture

Function

1. Improves texture
2. Prevents premature reaction

4. Epsom Salt (Magnesium Sulphate)

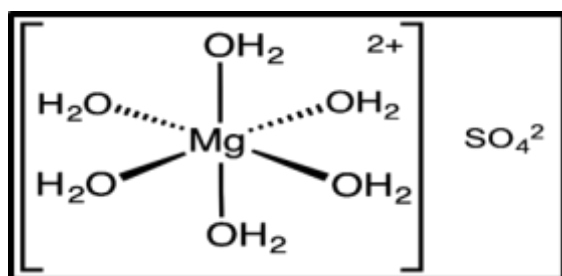


Fig 6.8 Structure of Magnesium Sulphate

Category

Bulking agent / Soothing agent.

Properties

1. Crystalline solid.
2. Water-soluble.

Function

1. Provides smooth texture
2. Soothes skin

5. Water

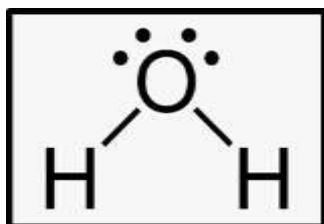


Fig 6.9 Structure of Water

Category

Binding agent.

Function

1. Helps in binding ingredients.
2. Assists in moulding.

6. Fragrance (Optional)

Category

Perfuming agent

Function

Improves product acceptability.

7. Kaolin

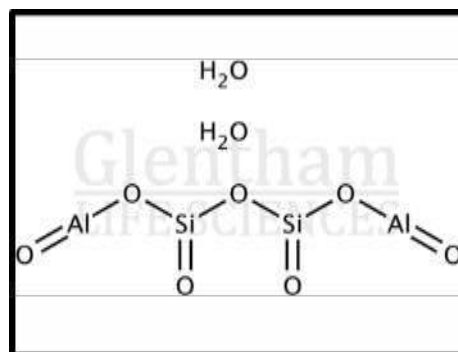


Fig 6.10 Structure of Kaolin

Category

Clay / Adsorbent.

Properties:

1. Fine white powder.
2. Absorbent in nature.
3. Smooth texture.
4. Insoluble in water.

Function:

1. Absorbs excess oil.
2. Removes impurities.
3. Gives cleansing effect.
4. Used in face packs and powders.

8. Polysorbate 80

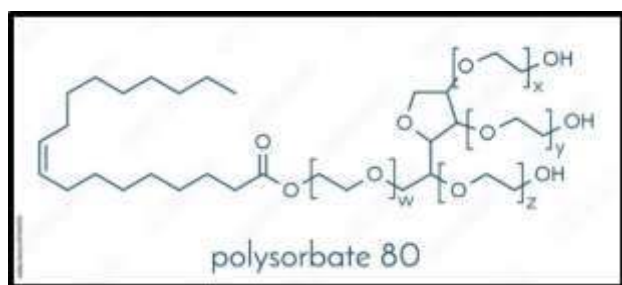


Fig 6.11 Structure of Polysorbate 80

Category

Non-ionic surfactant / Emulsifier.

Properties:

1. Yellow viscous liquid.
2. Soluble in water.
3. Stabilizes emulsions.
4. Mild in action.

Function:

1. Acts as emulsifying agent.
2. Helps mix oil and water.
3. Used as solubilizer.
4. Improves formulation stability.

9. Sodium Lauryl Sulphate

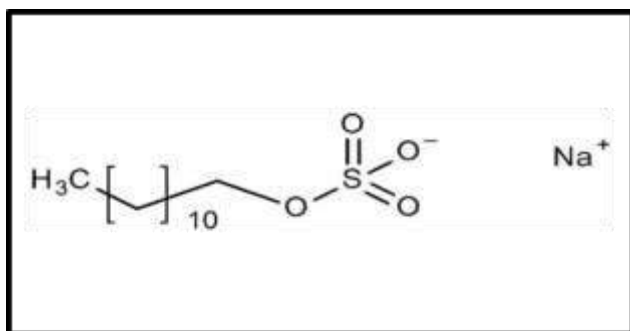


Fig. 6.12 Structure of Sodium lauryl sulphate

Category

Anionic surfactant / Detergent

Properties:

1. Good foaming ability.
2. Water soluble.
3. Strong cleansing action.
4. Surface active agent.

Function:

1. Produces foam.
2. Removes dirt and oil.
3. Used in shampoos and toothpaste.
4. Acts as cleansing agent.

7. METHODS

7.1 7.1 Extraction and Identification of Bioactive Compounds from Herbal Sources

7.1.1 Collection, Preparation and extraction of Neem Leaves: -

Procedure

1. Collection and Preparation of Neem Leaves:

1. Collect fresh neem leaves or seeds.
2. Wash thoroughly with distilled water to remove dust.
3. Dry under shade for 7–10 days until moisture is removed.
4. Grind the dried material into coarse powder using grinder.
5. Sieve the powder for uniform particle size.



Fig 7.1.1 Dried Neem Leaves and Powder Form

2. Weighing of Sample: -

Accurately weigh about 25–50 g of neem powder.

3. Preparation of Soxhlet Apparatus: -

1. Transfer neem powder into a cellulose extraction thimble.
2. Place the thimble inside Soxhlet extractor.

3. Fill round bottom flask with 250–500 mL ethanol.

Connect:

1. Round bottom flask
2. Soxhlet extractor
3. Condenser



Fig 7.1.2 Extraction of Neem extract

4. Extraction Process: -

1. Heat the apparatus using heating mantle.
2. Ethanol begins to boil and evaporate.
3. Vapours pass into condenser and condense into liquid.
4. Condensed solvent drips onto neem powder.

5. Bioactive compounds dissolve in ethanol.

6. When chamber fills, siphon arm transfers extract back into flask.
7. This cycle repeats continuously.

5. Extraction Time: -

Continue extraction for 6–8 hours or until solvent in siphon tube becomes colourless.

Identification Tests for Azadirachta Indica**1. Test for Alkaloids: -****Table No.7.1 Test of Alkaloids**

Test	Procedure	Observation	Inference
Dragendorff's Test	1. Prepare neem extract in dilute hydrochloric acid.2. Add Dragendorff's reagent.	Orange or reddish-brown precipitate appears.	Presence of alkaloids.
Mayer's Test	1. Take a small quantity of neem or garlic extract in a test tube.2. Add a few drops of dilute hydrochloric acid and filter if necessary.3. Add 2–3 drops of Mayer's reagent to the filtrate.	Cream-colored or white precipitate appears.	Presence of alkaloids is confirmed.
Wagner's Test	1. Take the plant extract in a test tube.2. Acidify with dilute hydrochloric acid.3. Add a few drops of Wagner's reagent.	Reddish-brown or brown precipitate forms.	Indicates the presence of alkaloids.

2. Test for Flavonoids**Table No.7.2 Test of Flavonoids**

Test	Procedure	Observation	Inference
Shinoda Test	Add magnesium turnings and concentrated hydrochloric acid to neem extract.	Pink or red coloration develops.	Presence of flavonoids.

3. Test for Glycosides**Table No.7.3 Test Of Glycosides**

Test	Procedure	Observation	Inference
Keller–Killiani Test	1. Add glacial acetic acid containing ferric chloride to the extract. 2. Carefully add concentrated sulphuric acid along the sides of the test tube.	Brown ring appears at the junction.	Presence of glycosides.

4. Test for Tannins**Table No. 7.4 Test of Tannins**

Test	Procedure	Observation	Inference
Ferric Chloride Test	Add a few drops of ferric chloride solution to neem extract.	Blue-black or green coloration appears.	Presence of tannins.

5. Test for Saponins**Table No.7.5 Test of Saponins**

Test	Procedure	Observation	Inference
Foam Test	Shake neem extract vigorously with water.	Persistent foam formation observed.	Presence of saponins.



6. Thin Layer Chromatography (TLC)

Table no.7.6 Test of TLC

Test	Procedure	Observation	Inference
Thin Layer Chromatography (TLC)	1. Prepare neem extract. 2. Spot the sample on a silica gel TLC plate. 3. Use a suitable solvent system such as toluene: ethyl acetate.	Distinct spots observed under UV light.	Confirms the presence of active constituents such as azadirachtin.

7.2.1 Extraction and Identification of Allium Sativum Extract

A) Collection and Preparation: -

The outer papery layers were removed, and the cloves were separated, thoroughly cleaned with distilled water to remove dirt and surface impurities, and then allowed to air-dry at room temperature. Once dried, the garlic cloves were crushed or coarsely ground using a mortar and pestle or mechanical grinder.

B) Cold Maceration Extraction: -

Fifty grams of crushed garlic were placed in a clean, dry conical flask, and ethanol was added in a 1:5 w/v ratio. The flask was sealed to prevent evaporation and kept in a cool, dark place for 3 to 7 days for maceration, with occasional stirring. Afterward, the mixture was filtered using Whatman filter paper, and the resulting extract was stored in amber-coloured bottles in cool, dry place or refrigerator to protect it from light and degradation.



Fig 7.2.1 Cold Maceration of Allium Sativum Extract

Identification Test for Allium Sativum

1. Sodium Nitroprusside Test

Table No.7.7 Test of sodium Nitroprusside

Test	Principle	Procedure	Observation	Inference
Sodium Nitroprusside Test	Sulfur-containing compounds like allicin react with sodium nitroprusside	1. Take garlic extract in a test tube.	Purple or violet coloration appears.	Presence of sulfur-containing compounds such as allicin is confirmed.

	in alkaline medium to produce color.	2. Add a few drops of sodium nitroprusside solution. 3. Add dilute ammonia solution.		
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2. Lead Acetate Test

Table No.7.8 Test of Lead Acetate

Test	Principle	Procedure	Observation	Inference
Lead Acetate Test	Sulfur compounds react with lead acetate to form dark-coloured lead sulphide precipitate.	1. Take garlic extract in a test tube. 2. Add a few drops of lead acetate solution.	Black or dark brown precipitate appears.	Indicates the presence of sulfur compounds including allicin.

3. Thin Layer Chromatography (TLC)

Table No. 7.9 Test of TLC

Test	Procedure	Observation	Inference
Thin Layer Chromatography (TLC)	1. Prepare garlic extract using ethanol. 2. Spot the sample on a silica gel TLC plate. 3. Use a suitable solvent system such as hexane: ethyl acetate. 4. Visualize under UV light or in an iodine chamber.	Distinct spot corresponding to allicin observed.	Confirms the presence of allicin in garlic extract.

4. High Performance Liquid Chromatography (HPLC)

Table No.7.10 Test of HPLC

Test	Principle	Procedure	Observation	Inference
High Performance Liquid Chromatography (HPLC)	Allicin is separated and identified based on retention time using HPLC.	Prepare garlic extract and inject into HPLC system; run using suitable mobile phase.	Characteristic peak obtained at specific retention time.	Confirms the presence and quantity of allicin.

UV-Visible Spectrophotometric Identification of *Azadirachta indica* and *Allium sativum*:-

UV-Visible spectrophotometry is a simple and reliable analytical technique used for identification of phytoconstituents present in herbal extracts. In *Azadirachta indica* (neem), the active constituent azadirachtin shows characteristic absorption peaks

in the UV region, mainly around 210–220 nm and 260–270 nm. For analysis, neem extract is prepared using ethanol or methanol and scanned in the range of 200–400 nm using a UV-Visible spectrophotometer. The appearance of specific absorption peaks confirms the presence of azadirachtin and related bioactive compounds



responsible for insecticidal and anti-tick activity (Biswas et al., 2002).

Similarly, allicin present in garlic (*Allium sativum*) can also be identified using UV–Visible spectrophotometry. Garlic extract prepared in ethanol exhibits a characteristic absorption peak around 240 nm due to sulfur-containing compounds present in allicin. The obtained peak confirms the presence of allicin, which is responsible for antimicrobial, antiparasitic, and insect-repellent properties (Ankri & Mirelman, 1999). Thus, UV–Visible spectrophotometry serves as a rapid, economical, and effective method for identification and standardization of neem and garlic extracts used in herbal anti-tick formulations.

Method of Preparation of Herbal Anti-Tick Bath Bomb: -



Formulation of Herbal Anti-Tick Bath Bomb using *Azadiracta Indica* and *Allium Sativum*

Batches Prepared: - Batch 1: - Failed

Table 7.11 F1

Sr. No	Ingredients	Quantity	Uses
1	Sodium Bicarbonate	5 gm	Weak Base
2	Citric Acid	4 gm	Strong Acid
3	Corn Starch	2.5 gm	Binder
4	Magnesium Sulphate	2.5 gm	Muscle Relaxant
5	Polysorbate 80	2 ml	Emulsifier
6	Kaolin	1.5 gm	Hardening Agent
7	SLS	0.1 ml	Surfactant
8	Neem Extract	1 ml	Antibacterial
9	Garlic Extract	1 ml	Antimicrobial
10	Essential Oil	q.s	Fragrance

Batch 2: - Failed

Table 7.12 F2

Sr. No	Ingredients	Quantity	Uses
1	Sodium Bicarbonate	5 gm	Weak Base
2	Citric Acid	2.5 gm	Strong Acid
3	Corn Starch	1 gm	Binder
4	Magnesium Sulphate	2.5 gm	Muscle Relaxant
5	Polysorbate 80	2 ml	Emulsifier
6	Kaolin	1.5 gm	Hardening Agent
7	SLS	0.1 ml	Surfactant
8	Neem Extract	1 ml	Antibacterial
9	Garlic Extract	1 ml	Antimicrobial
10	Essential Oil	q.s	Fragrance

Batch 3: - Failed



Table 7.13 F3

Sr. No	Ingredients	Quantity	Uses
1	Sodium Bicarbonate	5 gm	Weak Base
2	Citric Acid	2.5 gm	Strong Acid
3	Corn Starch	2.5 gm	Binder
4	Magnesium Sulphate	2.5 gm	Muscle Relaxant
5	Polysorbate 80	2 ml	Emulsifier
6	Kaolin	-	Hardening Agent
7	SLS	0.1 ml	Surfactant
8	Neem Extract	1 ml	Antibacterial
9	Garlic Extract	1 ml	Antimicrobial
10	Essential Oil	q.s	Fragrance

Batch 4: - Passed

Table 7.14 F4

Sr. No	Ingredients	Quantity	Uses
1	Sodium Bicarbonate	6.7 gm	Weak Base
2	Citric Acid	3.3 gm	Strong Acid
3	Corn Starch	3.3 gm	Binder
4	Magnesium Sulphate	3.3 gm	Muscle Relaxant
5	Polysorbate 80	2.7 ml	Emulsifier
6	Kaolin	2 g	Hardening Agent
7	SLS	0.13 ml	Surfactant
8	Neem Extract	1.3 ml	Antibacterial
9	Garlic Extract	1.3 ml	Antimicrobial
10	Essential Oil	q.s	Fragrance

8. EVALUATION

8.1.1 Identification test for Azadirachta Indica

8.1 Identification Test for Azadirachta Indica and Allium Sativum

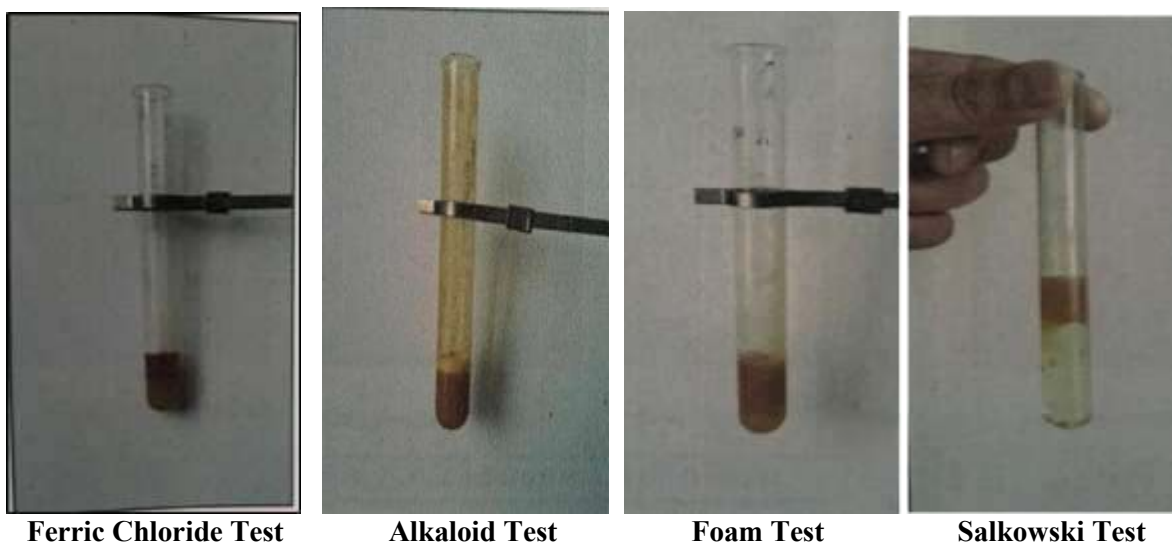
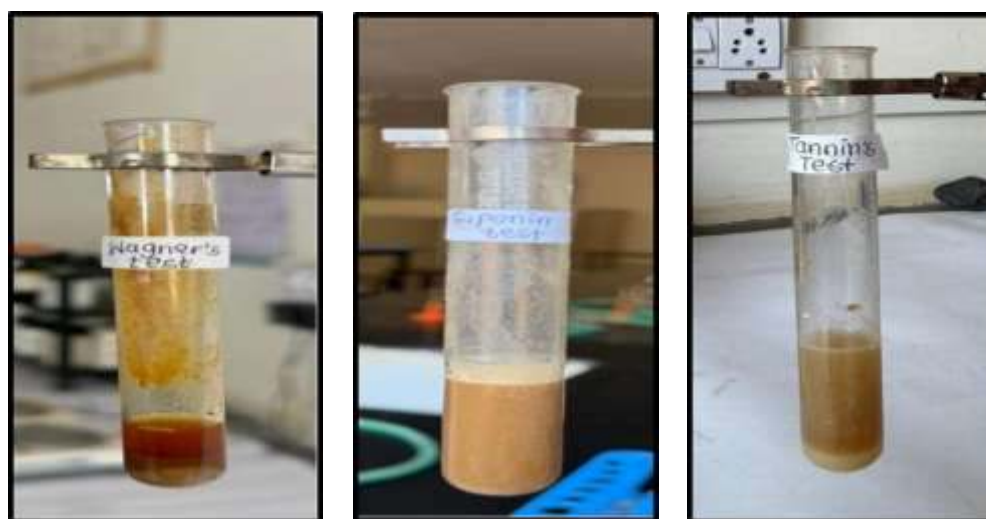


Fig.8.1.1 Identification Test for Azadirachta Indica

Table No. 8.1.1 Result of the Identification Test of Azadirachta Indica

Test Name	Reagent Used	Observation	Result
Ferric Chloride Test	Ferric chloride solution (FeCl ₃ , 1%)	Dark green / blue-black coloration	Presence of phenolic compounds / tannins
Alkaloid Test	Dragendorff's reagent	Orange-red precipitate	Presence of alkaloids
Foam Test	Distilled water	Stable foam formation	Presence of saponins
Salkowski Test	Chloroform + Concentrated H ₂ SO ₄	Reddish-brown interface	Presence of terpenoids / steroids
Shinoda Test	Magnesium ribbon + Concentrated HCl	Pink / red coloration	Presence of flavonoids

8.1.2. Identification test for Allium Sativum



Wagner's Test Foam Test Ferric Chloride Test
 Fig.8.1.2 Identification test for Allium Sativum

Table No.8.1.2 Result of Identification Test of Allium Sativum

Test Name	Phytochemical Detected	Procedure	Positive Observation
Wagner's Test	Alkaloids	Add a few drops of Wagner's reagent (iodine in potassium iodide) to the plant extract.	Reddish-brown or brown precipitate forms.
Foam Test	Saponins	Shake the extract vigorously with water in a test tube for 1–2 minutes.	Stable persistent froth/foam appears for several minutes.
Ferric Chloride Test	Tannins	Add a few drops of 5% ferric chloride solution to the extract.	Blue-black or greenish-black coloration appears.

8.1.3. UV-Visible Spectrophotometric Study

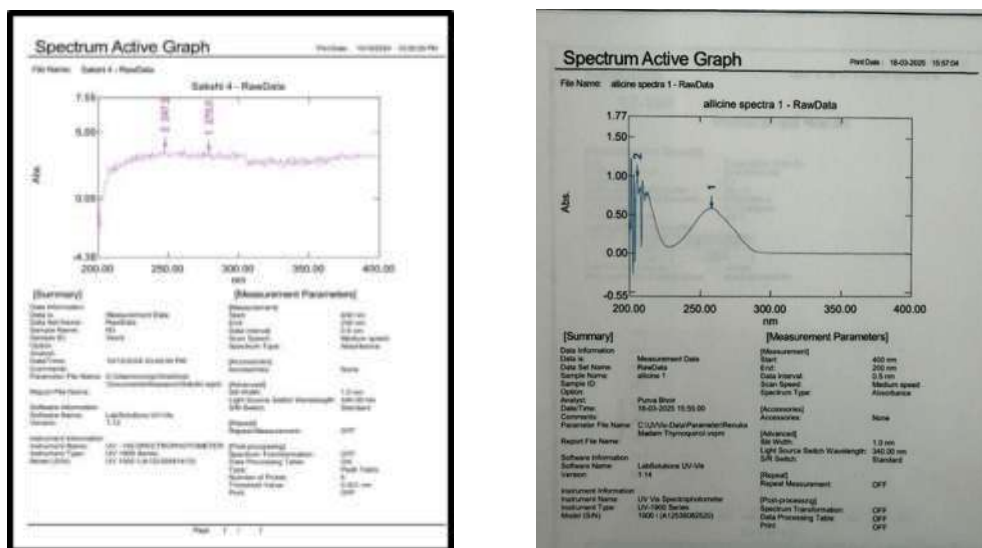


Fig.8.1.3 UV-Visible Spectrophotometric Stud

Table 8.1.3 UV-Visible Spectrophotometric Study

Parameter	Allium Sativum	Azadirachta Indica
Spectrum appearance	Smooth with clear peaks	Broad, noisy, almost flat
Main peaks observed	~205–215 nm and ~255–260 nm	Weak peaks near 247 nm and 279 nm
Maximum absorbance	~1.2 AU	~3–4 AU
Baseline behaviour	Stable and near zero after 300 nm	Elevated baseline throughout
Peak clarity	Well-defined peaks	Poorly defined peaks
Noise level	Low	High
Data quality	Good and reliable	Poor/unreliable
Detector condition	Within working range	Likely detector saturation
Likely issue	None/minor	Concentration too high, turbidity, or baseline error
Recommended action	Can be used for analysis	Dilute sample and rescan
Overall conclusion	Valid UV–Vis spectrum	Invalid or poor-quality spectrum

8.2. Evaluation Test For Azadirachta Indica and Allium Sativum

1. Weight variation test

Procedure

- Select 10 bath bombs randomly from the prepared batch.
- Clean the surface of bath bombs to remove loose powder.
- Weigh each bath bomb individually using digital weighing balance.

- Record the individual weights.
- Calculate the average weight of all bath bombs.
- Determine the deviation of each bath bomb from average weight.

Formula

Percentage Weight Variation =

$$\frac{\text{Individual Weight} - \text{Average Weight}}{\text{Average Weight}} \times 100$$

Bath Bomb No.	Individual Weight (g)
1	10.00
2	9.900
3	9.910
4	10.00
5	9.840
6	8.740
7	9.830
8	10.00
Average	9.778



Fig.8.2.1 Weight Variation Test

2. Friability Test

Procedure

- Select clean and completely dried bath bombs.

- Remove loose powder or dust from the surface using soft brush.
- Accurately weigh the bath bombs and record the initial weight as W_1 .
- Place the bath bombs in Roche friabilator.
- Rotate the friabilator at 25 rpm for 4 minutes (100 revolutions).
- Remove the bath bombs carefully after completion of test.
- Remove loose dust from the surface again.
- Reweigh the bath bombs and record the final weight as W_2 .

Formula

Where:

W_1 = Initial weight of bath bombs

W_2 = Final weight after friability test

$W_1 = 10.0$ $W_2 = 9.9$



Fig 8.2.2 Friability Test

3. Foamability test

Procedure

- Take 100 mL of distilled water in a measuring cylinder.

- Add approximately 1 g of crushed bath bomb into the cylinder.
- Shake the cylinder vigorously for 10 minutes.
- Allow the solution to stand for 1–5 minutes.
- Measure the height of foam formed above the liquid surface.
- Record foam height immediately and after standing time to evaluate foam stability.

8.2.1 Observation Table of foamability

Parameter	Observation
Initial foam height	5 cm
Foam height after 5 min	3 cm
Nature of foam	Stable
Foam appearance	Fine



Fig 8.2.3 Foamability Test

5. Dissolution test

Procedure

- Take 200 mL of distilled water in a clean beaker.
- Maintain water temperature at approximately 25–37°C.
- Place one bath bomb gently into the water.
- Start the stopwatch immediately.
- Observe fizzing action, disintegration, and dissolution behaviour.
- Record the time required for complete dissolution of the bath bomb.
- Observe whether the solution becomes clear or uniformly dispersed.

8.2.2 Observation Table of dissolution test

Parameter	Observation
Water temperature	32 °C
Complete dissolution time	3 min
Nature of dissolution	Uniform
Residue present	No



Fig 8.2.4 Dissolution Test

6. Moisture content determination

Procedure

- Take a clean and dry porcelain dish and weigh it accurately. Record the weight as W_1 .
- Crush the bath bomb into fine powder.
- Place about 2–5 g of powdered sample into the dish.
- Weigh the dish containing sample and record the weight as W_2 .
- Place the dish in a hot air oven maintained at 105°C for about 2–3 hours.
- Remove the dish carefully and cool it in a desiccator.
- Reweigh the dried sample and record the final weight as W_3 .
- Repeat the drying process until constant weight is obtained.

Formula

Where:

W_1 = Weight of empty dish

W_2 = Weight of dish + sample before drying

W_3 = Weight of dish + sample after drying

$W_1 = 24.55$

$W_2 = 26.88$

$W_3 = 28.20$

Moisture content is less than 5 percent.



Fig. 8.2.5 Moisture Content Test

7. Disintegration test

Procedure

- Take 200 mL of distilled water in a clean beaker.
- Maintain the temperature of water at approximately $25\text{--}37^\circ\text{C}$.
- Place one bath bomb gently into the water.
- Start the stopwatch immediately.
- Observe the bath bomb carefully for breaking, fizzing, and disintegration.
- Record the time required for complete disintegration of the bath bomb into fine particles.
- Note whether any residue remains after disintegration.

8.2.3 Observation Table of disintegration test

Parameter	Observation
Water temperature	32°C
Complete disintegration time	3 min/sec
Residue present	No
Nature of disintegration	Uniform



Fig. 8.2.6. Disintegration Test

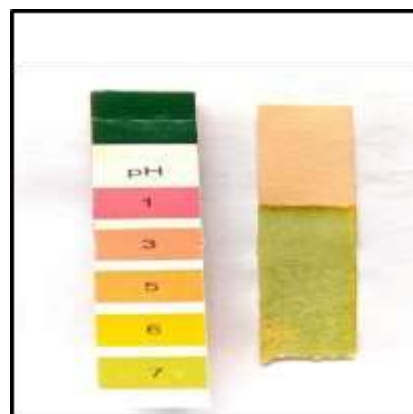


Fig 8.2.7 PH Test

7. PH determination

Procedure

- Take 1 g of crushed bath bomb in a beaker.
- Add 100 mL of distilled water to prepare the solution.
- Stir the solution properly using glass rod until complete dissolution.
- Calibrate the pH meter using standard buffer solutions if required.
- Dip the electrode of pH meter into the prepared solution.
- Allow the reading to stabilize and record the pH value.
- If pH paper is used, compare the colour developed with standard colour chart.

8.2.4 Observation Table of pH test

Parameter	Observation
Quantity of sample	2 g
Volume of water	100 mL
Observed pH	7

8. Effervescent test

Procedure

- Take 200 mL of distilled water in a clean beaker.
- Maintain water temperature at approximately 25–37°C.
- Place one bath bomb gently into the water.
- Start the stopwatch immediately.
- Observe the release of bubbles and fizzing action.
- Record the time required for complete effervescence and dissolution of the bath bomb.
- Observe the intensity and uniformity of fizzing.

8.2.5 Observation Table of Effervescence test

Parameter	Observation
Water temperature	32 °C
Complete effervescence time	2 min/sec
Nature of fizzing	Strong
Dissolution	Complete



Fig. 8.2.8 Effervescent Test

8.3 Final Formulated Bath Bomb:

Herbal Anti-tick bathbomb using Azadirachta Indica and Allium Sativum extract formulated successfully.



Fig 8.3.1 Final Formulated Bath Bomb

9. RESULT

Sr. No.	Parameter	Result
1	Weight Variation Test	Average weight found to be 9.778 g; weight variation within acceptable range except slight deviation in sample 6
2	Friability Test	Initial weight = 10.0 g; Final weight = 9.9 g; friability found within acceptable limit
3	Foamability Test	Initial foam height = 5 cm; foam height after 5 min = 3 cm; foam was stable and fine
4	Dissolution Test	Complete dissolution observed within 3 min at 32°C; dissolution was uniform with no residue
5	Moisture Content Determination	Moisture content found to be less than 5%
6	Disintegration Test	Complete disintegration observed within 3 min/sec at 32°C; no residue remained
7	pH Determination	pH of bath bomb solution was found to be 7, indicating neutral nature
8	Effervescent Test	Strong and uniform effervescence observed; complete effervescence within 2 min/sec

CONCLUSION

The herbal anti-tick bath bomb formulated using neem (*Azadirachta indica*) and garlic (*Allium sativum*) was successfully developed and



evaluated. The formulation showed good physical properties, stability, and acceptable pH suitable for skin compatibility. Evaluation studies confirmed uniform weight, good mechanical strength, controlled moisture content, and effective effervescence with proper dissolution and disintegration behaviour. The product also exhibited good foam ability and demonstrated effective anti-tick activity along with being safe and non-irritant. Overall, the study concludes that the developed bath bomb is an eco-friendly, effective, and promising alternative to synthetic anti-tick products for veterinary use.

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