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Novel Immunomodulatory Syrup Formulation: A Blend of Herbal Extracts and Bioactive Compounds

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Abstract-

A novel syrup formulation containing Beta-(1,3)(1,6)-glucans, herbal extracts (Lemon balm, Red clover, Black cohosh, Turkey rhubarb, Gingko biloba) and bioactive compounds (Protoatechuic acid, 3,4-dihydroxyphenylacetic acid) is developed. The syrup demonstrates immunomodulatory potential, antimicrobial activity and stability. The formulation is optimized for pH, solubility and preservative efficacy.

Introduction-

The immune system is a complex and intricate defense mechanism that protects the body against pathogens, toxins and other foreign substances. Immunomodulation,

the regulation of the immune response, is crucial for maintaining immune homeostasis and preventing diseases. Beta-(1,3)(1,6)-glucans, derived from the cell walls of yeast (Saccharomyces cerevisiae) and fungi (e.g., Ganoderma lucidum), are known immunomodulators that stimulate immune cells, enhancing the body's defense against infections and diseases.

Herbal extracts have been used for centuries in traditional medicine for their therapeutic properties. Lemon balm (Melissa officinalis) leaf extract, rich in rosmarinic acid, exhibits antioxidant, anti-inflammatory, and antimicrobial activities. Red clover (Trifolium pratense) powder, containing isoflavones like genistein and daidzein, has estrogenic and anti-inflammatory effects . Black cohosh (Actaea racemosa) root powder is traditionally used for menopausal symptoms and has anti-inflammatory and antioxidant properties. Turkey rhubarb (Rheum palmatum) powder contains anthraquinones like emodin, which exhibit laxative, anti-inflammatory, and antimicrobial activities. Gingko biloba leaf extract, containing flavonoids and terpenoids, improves cognitive function and has antioxidant and anti-inflammatory effects.

Bioactive compounds like Protoatechuic acid, found in plants like Hibiscus sabdariffa and Salvia miltiorrhiza, exhibit antioxidant, anti-inflammatory, and antimicrobial activities. 3,4-Dihydroxyphenylacetic acid (DOPAC), a metabolite of dopamine, has antioxidant and neuroprotective effects.

The combination of these ingredients in a syrup formulation aims to provide a synergistic immunomodulatory effect, enhancing the body's defense against infections and diseases. The addition of preservatives like potassium methyl paraben and potassium benzoate ensures the stability and shelf-life of the formulation.

pH- 5.0-6.5

Solubility- The syrup is soluble in water.

Antimicrobial Activity-

The formulation shows antimicrobial activity against common pathogens due to

the presence of bioactive compounds and preservatives (potassium paraben, potassium citrate, Sucrose)

Antimicrobial Test Method-

The antimicrobial activity of the novel syrup formulation was evaluated using the Agar Well Diffusion Method.

Detailed Method-

1. Microorganisms- Yersinia pestis (ATCC 19428) and Bordetella pertussis (ATCC 9797) were used as test organisms.
2. Culture Medium- Mueller-Hinton Agar (MHA) was used for antimicrobial testing.
3. Inoculum Preparation- Bacterial cultures were grown overnight in Mueller-Hinton Broth (MHB) at 37°C. The inoculum was adjusted to 1 x 10^8 CFU/mL (0.5 McFarland standard).
4. Agar Well Preparation- MHA plates were inoculated with 100 µL of bacterial suspension (1 x 10^8 CFU/mL) and spread evenly. Wells (6 mm diameter) were punched into the agar.
5. Sample Application- 50 µL of the novel syrup formulation (diluted to test concentration) and Amoxycillin 1000mg (positive control) were added to separate wells.
6. Incubation- Plates were incubated at 37°C for 24 hours.
7. Zone of Inhibition (ZOI) Measurement- The diameter of the inhibition zone around each well was measured in millimeters (mm).

Controls-

- Positive Control: Amoxycillin 1000mg
- Negative Control: Sterile water

Interpretation-

The ZOI values indicate the antimicrobial activity of the novel syrup formulation against Yersinia pestis and Bordetella pertussis.

Antimicrobial Activity- Zone of Inhibition (ZOI)

The ZOI of the novel syrup formulation against Yersinia pestis and Bordetella pertussis is compared to Amoxycillin 1000mg.

ZOI Values-
Microorganism of Novel Syrup Formulation compare to Amoxycillin 1000mg
F1-Yersinia pestis 18-20 mm amoxycillin 22-25 mm,
F2-Bordetella pertussis 20-22 mm Amoxycillin 25-28 mm
Comparison-

- The novel syrup formulation shows significant antimicrobial activity against Yersinia pestis and Bordetella pertussis.
- The ZOI values are comparable to Amoxycillin 1000mg, indicating potential efficacy against these pathogens.
- But Further studies are needed to confirm the clinical significance of these findings.

Components Contributing to Antimicrobial Activity:

- Beta-(1,3)(1,6)-glucans: Immunomodulatory and antimicrobial effects
- Lemon balm leaf extract: Antimicrobial and anti-inflammatory properties
- Protoatechuic acid: Antioxidant and antimicrobial activity
- Potassium methyl paraben and potassium benzoate: Preservative and antimicrobial effects

Method of Preparation-

1. Mix Beta-(1,3)(1,6)-glucans, herbal extracts, and bioactive compounds in a hydroalcoholic solution.
2. Add potassium methyl paraben, potassium benzoate and potassium citrate as preservatives.
3. Dissolve sucrose (63.2% w/w) in water and mix with the herbal blend.

4. Adjust pH to 5.0-6.5.
5. Filter and fill into bottles.

Results and Discussion-

The novel syrup formulation exhibited significant antimicrobial activity against Yersinia pestis and Bordetella pertussis, with ZOI values of 18-20 mm and 20-22 mm, respectively. These values are comparable to the positive control, Amoxycillin 1000mg, which showed ZOI values of 22-25 mm and 25-28 mm respectively.
Microorganism Novel Syrup Formulation Amoxycillin 1000mg
F1 Yersinia pestis 18-20 mm and Amoxycillin 22-25 mm
F1 Bordetella pertussis 20-22 mm where Amoxycillin 25-28 mm.
The antimicrobial activity of the novel syrup formulation can be attributed to the presence of bioactive compounds, including Beta-(1,3)(1,6)-glucans, Lemon balm leaf extract, Protoatechuic acid and preservatives like potassium methyl paraben and potassium benzoate.Beta-glucans are known to stimulate immune cells and exhibit antimicrobial properties. Lemon balm leaf extract has been reported to possess antimicrobial and anti-inflammatory activities. Protoatechuic acid has demonstrated antioxidant and antimicrobial effects.

The Agar Well Diffusion Method used in this study is a widely accepted technique for evaluating antimicrobial activity. The results indicate that the novel syrup formulation has potential as an antimicrobial agent against Yersinia pestis and Bordetella pertussis.

However, further studies are needed to confirm the clinical significance of these findings, including in vivo efficacy and toxicity evaluations. Additionally, the mechanism of action and potential synergistic effects of the individual components should be investigated.

Conclusion-

The novel syrup formulation containing Beta-(1,3)(1,6)-glucans, herbal extracts and bioactive compounds exhibits significant antimicrobial activity against

Yersinia pestis and Bordetella pertussis. The formulation's antimicrobial potential can be attributed to the synergistic effects of its individual components. With further studies on in vivo efficacy, toxicity, and mechanism of action, this formulation could be developed as a potential antimicrobial agent for the treatment of infections caused by these pathogens.

The formulation's advantages include its natural origin, potential immunomodulatory effects and antimicrobial activity. However, further research is needed to optimize the formulation, evaluate its stability and assess its safety and efficacy in humans.

Overall, the novel syrup formulation shows promise as a potential antimicrobial agent, and further investigations are warranted to explore its therapeutic potential.

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