EFFECTIVENESS OF *MORINGA OLEIFERA* EXTRACT COMBINED WITH PEG 400 AGAINST NON-SPECIFIC BACTERIA

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ABSTRACT

The ever-increasing threat of antibiotic-resistant bacteria has led to a high demand for new solutions and agents that decreases and slows down this threat. Although powerful antibiotics exist in the field of medicine, its improper and irresponsible use has led to an increase in resistant strains of previously curable bacteria. One existing solution used by many studies is the use of Moringa oleifera on resistant strains of bacteria. However, developmental research on enhancing the antibacterial properties of M. oleifera is seldom focused on. This suggests a need for innovative ideas to improve its antibacterial properties and in turn, contribute to the beneficiaries of this study. This study aims to determine the synergistic effect of the osmotic compound PEG-400 on the Antibacterial properties of Moringa in terms of the diameter of the zone of inhibition. This study utilized 5 g of coarsely grinded leaves of M. oleifera mixed with 100 ml of distilled water. Afterward, the mixture was put through a process of hot water extraction and centrifugation to obtain the extract. Moreover, this study used five large Petri dishes containing one replication of each M. oleifera extract disc, M. oleifera extract with PEG 400 disc, and dry disc. The results of this study were obtained using Antimicrobial Susceptibility Test (AST). The researchers concluded that M. oleifera extract combined with PEG 400 does not exhibit an increased activity and that PEG 400 does not have a synergistic effect on the antibacterial properties of M. oleifera.

INTRODUCTION

As stated by Vidyagasar (2019), in the United States alone, 23,000 people die from antibiotic-resistant bacteria. A discovery of Cykanzski (2018) highlights the need for antibacterial alternatives and improvement. Throughout the years, due to the constant evolution of antibiotic resistance in bacteria, issues in the medical area has occurred. Oftentimes, the reason behind this is the misuse of antibiotics, the wrong dosage, or the antibiotics is simply inefficient. Hence, the need to search for viable treatments that can inhibit these antibiotic-resistant bacteria constantly. This research aims to create a viable solution for the inhibition of bacteria.

Accordingly, with previous extensive research, Shedoeva, et al, 2019 discovered that many medical plants synthesize equivalent or closely related compounds and *Moringa oleifera* was the focus of researchers. *M. oleifera*, has long been proven to exhibit a low minimum inhibitory concentration at values of 16 micrograms/mL on some bacteria. Additionally, *M. oleifera* extracts at a concentration of 256 micrograms/mL inhibits all bacterial growth. So, in this research researchers aim to improve the antibacterial properties of *M. oleifera*, with the use of PEG 400 which in Chirife 1983 was proven to have antibacterial properties at a 100% concentration compound and has recently been proven to have an existing synergistic effect on existing antibiotics. However, studies connected to the use of PEG 400 osmotic compounds for improving the antibacterial efficacy of *M. oleifera* are in the absence of any osmotic compound, for that matter, as a method of enhancing the antibacterial effectiveness of Moringa oleifera.

METHODOLOGY

This study employed a static group experimental research design that examined the effect of PEG 400 on the antibacterial properties of Moringa oleifera leaf extract against a non-specific wound-swabbed bacteria.

Materials and Methods

The leaves were shade-dried for one (1) week. The dried leaves were then coarsely-grinded using pestle and mortar to obtain powder. Afterwards, five (5) grams of the powder was mixed with 100 mL of distilled water and placed atop a water bath at 40 degrees Celsius for 1 hour. Extracts were filtered into a container and were placed in a beaker to repeat the process for 30 minutes. The extracts were once again filtered and placed in 5 mL tubes for centrifugation. The extracts were centrifuged for five (5) minutes. The supernatant was set aside and was used to impregnate the filter paper disks in a Petri dish. The researchers first produced the extract of the plant that was used to impregnate filter paper disks for a total of 24 hours. The surgery ward in Tarlac Provincial Hospital provided bacteria swabbed from a wound stored in a tube containing 1 mL of Normal Saline Solution (NSS). The researchers then stored the bacteria at 4 degrees Celsius. Nutrient agar was mixed with 300 mL of distilled water and was heated to a boil until the solution was homogeneous. Then 15 mL of blood was collected from one of the researchers and was added to the Blood Agar Media. It was then shook rhythmically to remove bubbles on the surface.

A syringe was used to drop the bacterial solution to a Petri dish and was streaked onto Blood Agar Media using sterile inoculating loop. The Petri Dish was incubated for 24 hrs. at 37 degrees Celsius. A pure colony was selected and inoculated in a tube and was then incubated for 16 hours at 37 degrees Celsius. The resulting subculture was compared with a 0.5 McFarland Std. and was swabbed onto five (5) Petri dishes containing BAM. The corresponding filter paper disks were placed on the surface of the agar plate and were incubated for 20 hours.

Preparation of Experimental and Control Groups

This study made use of five (5) Petri dishes containing one replication of each M. oleifera extract discs, M. oleifera extracts with PEG 400 discs, and dry discs. Setup A was prepared using 20 mL of standard extract and 10 mL of PEG 400 compound, while setup B made use of 30 mL of the standard extracts.The negative control or the dry discs were labeled as setup C.

Statistical Treatment

The results of this study were obtained using the disc diffusion method in the laboratory for antimicrobial susceptibility testing.This study made use of t-test and ANOVA single factor. The zone of inhibition of setup A and setup B were tested for their significant difference with t-test of two sampled means. The use of ANOVA single factor provided an overall result on the significant difference among all existing setups.

RESULTS

PETRI DISH	Setup A	Setup B	Negative Control
1	0.000011	0.00001	0
2	0.00000	0.00000	0
3	0.00000	0.00000	0
4	0.00000	0.00000	0
5	0.00000	0.00000	0

VARIABLE	MEAN SCORE
Setup A (Moringa oleifera extract with PEG 400)	0.0000022
Setup B (Moringa oleifera extract only)	0.000002

t-computed value = 1

t-critical value = 2.78

Probability = 0.3739

SETUPS	MEAN INHIBITION
Setup A (Moringa oleifera extract with PEG 400)	0.0000022
Setup B (Moringa oleifera extract only)	0.000002
Setup C (Negative control)	0

F-computed value = 0.51

F-critical value = 3.89

Probability = 0.6173

CONCLUSION

Due to the evolution of antibiotic-resistant bacteria, this study intended to provide a more effective antibacterial solution made up of M. oleifera extract and PEG 400 that inhibits a non-specific bacterium from an infected wound. This study also serves as a prerequisite to further research focusing on the development of herbal ointments.

As was discussed in the findings of this research, setup A did not exhibit an increase in bactericidal activity leading us to conclude that PEG 400 does not have a synergistic effect on the antibacterial properties of M. oleifera and accept the null hypothesis. However, it is unknown whether PEG 400 decreased or simply did not affect the antibacterial properties of the extract.

Moreover, the factors that reduce the substance of this research are non-standard methodological procedures in disk impregnation as well as the lack of samples of known bacterial isolates since the researchers were unable to find studies that combine M. oleifera with PEG compounds. Therefore, impeding disk impregnation procedures. The methods of introducing the compound to the extract is unstandardized and the ratio of compound to extract was unknown. Meanwhile, this study was unable to make use of bacterial isolates due to the restrictions caused by the pandemic on Covid-19 pandemic referral hospitals.

AFFILIATION

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