Comparative Study of Antimicrobial Natural Products in Traditional Medicine Plants

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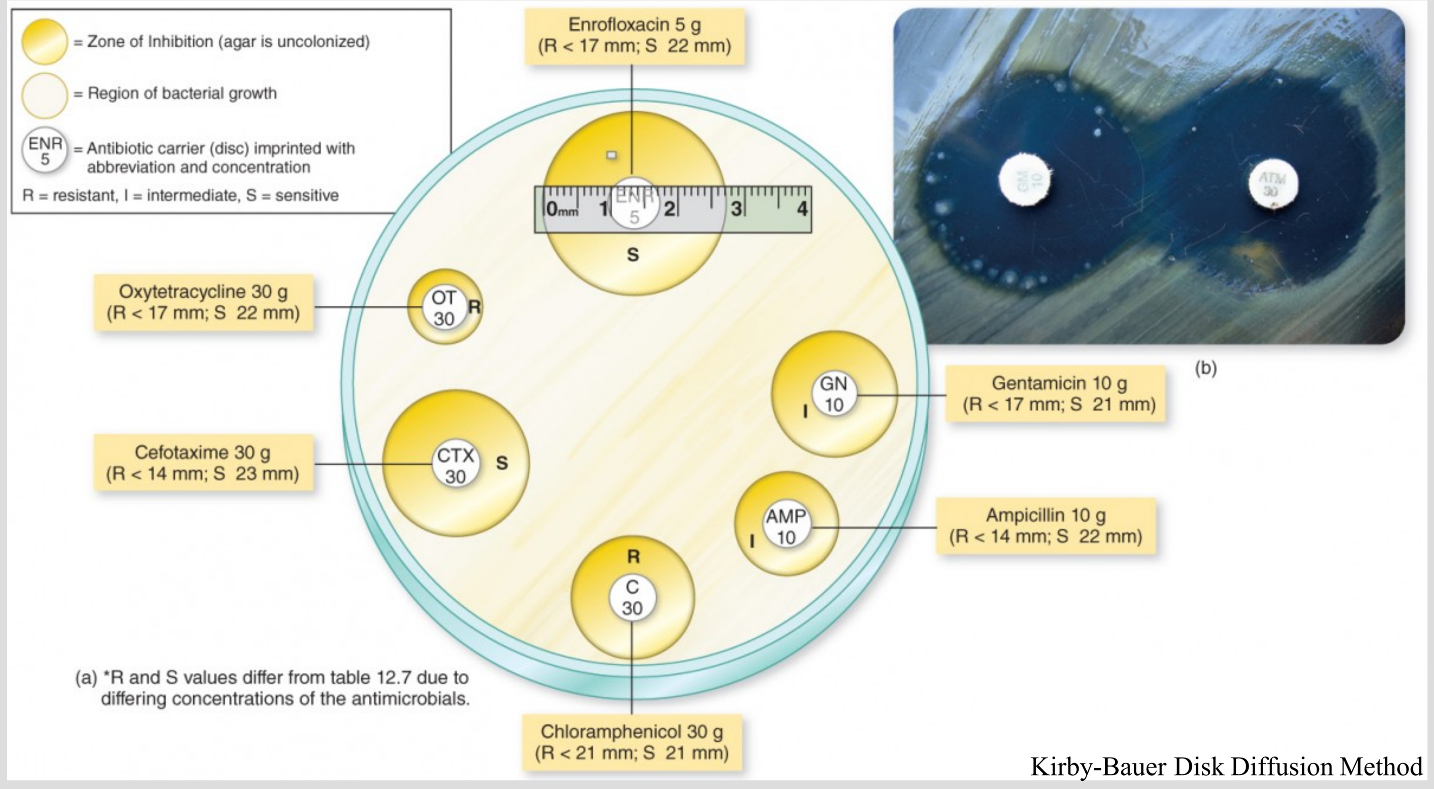
INTRODUCTION

In spite of the increasing progress made in the microbiology area, infectious diseases are still a significant problem worldwide. The need for the development of more effective and safe antimicrobial agents has therefore stimulated investigations focused on natural products as source of new leading antimicrobial drugs. Currently, such drugs are widely developed and utilized in the traditional eastern and western medicine. Traditional eastern and western medicine has a long history as it has been used over thousands of years for the prevention and treatment of various diseases. In recent times, it is making a rapid progress in scientific investigation and attracting great attention due to its antimicrobial properties. However, not all of the medicine used in these areas are widely understood. As such, we investigated on the characterization of novel products in specific traditional eastern medicinal plants and their western counterparts; and the effectiveness of their antimicrobial properties. The western counterparts of the traditional eastern medicinal plants were chosen based on the same type of symptoms they treat. Through the use of antimicrobial assays and high-performance liquid chromatography, specific metabolic compounds contained in the plants were identified and analyzed. The antimicrobial properties of the natural products found in traditional eastern and western medicinal plants were compared using the Kirby-Bauer disk diffusion method. This information and the comparatively favorable compounds could be used to develop more natural, effective medications in order to help remedy the problem of bacterial and infectious disease.

Western (Native American) Medicinal Plant	→ Symptom Treated ←	Eastern (Chinese) Medicinal Plant
Coffeeberry Tree Bark	→ Pain ←	Cinnamon Bark
Manzanita Tree Bark	→ Urinary Issues ←	Chinese Yam
Western Redbud Shoot Bark	→ Digestive Issues ←	Chinese Ginseng

MATERIALS & METHODS

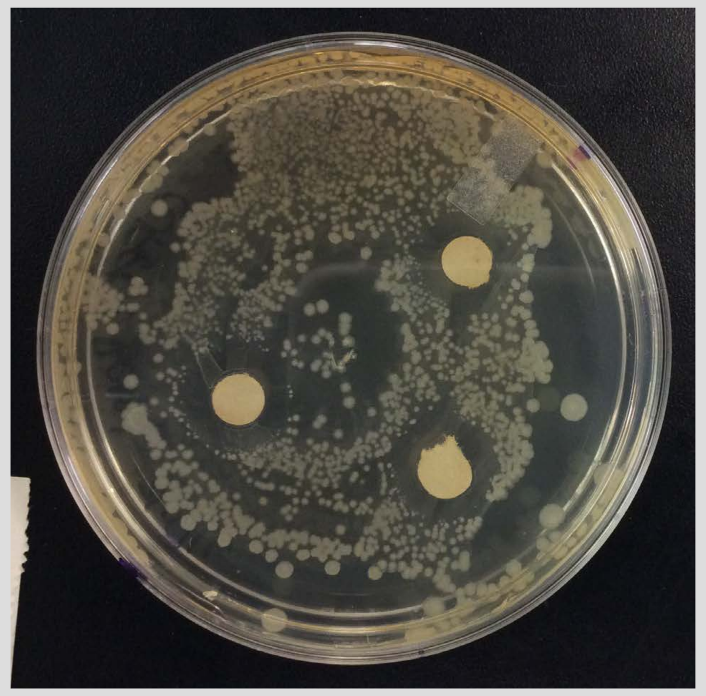
Dried Chinese traditional medicine plant samples were purchased, while Western traditional medicine plant samples were obtained from a garden. All samples were reduced to powders using scissors and razor blades or mortar and pestle. Their essences were extracted using an extraction kit or sonicator and reflux. The extraction kit entailed dissolving the samples in 300 μL of a mixture of 5 μL Protease Inhibitor Cocktail and 995 μL Plant Extraction Buffer, homogenizing the mixtures with a pestle, chilling on ice for 10 minutes, and centrifuging at 4° C for 5 minutes. The other method involved dissolving the samples in 96% methanol, sonicating for 1 hour at 25° C, reflux for 4 hours at 70° C, removing methanol through a vacuum in the AutoVac, then rehydrating the extract with 5mL of 50% methanol. The produced essences were then tested of their antimicrobial properties.
LB agar plates were prepared by mixing 35g of LB agar powder with 500 mL of distilled water, cooling the solution, and pouring it into petri dishes to solidify. Additionally, bacteria solutions were created by adding 3 colonies of E.coli bacteria to LB broth and leaving to reproduce. Once the plates and bacteria solutions were prepared, the antimicrobial assays were performed using the Kirby-Bauer disk diffusion method. 6mm hole punches of filter paper were created and sterilized in the Autoclave machine. Next, 50 μL of bacteria were placed on the surface of the plates and spread using a spreader. Three paper disks were placed separately on each plate, then 15 μL of plant extract was pipetted onto the disks. Three plates were created for each plant sample, plus one positive and one negative control. The positive control was 1 mg/mL ampicillin, and the negative control was de-ionized water. Finally, the plates were incubated for 24 hours at 37° C, then removed and their zones of inhibition were measured.



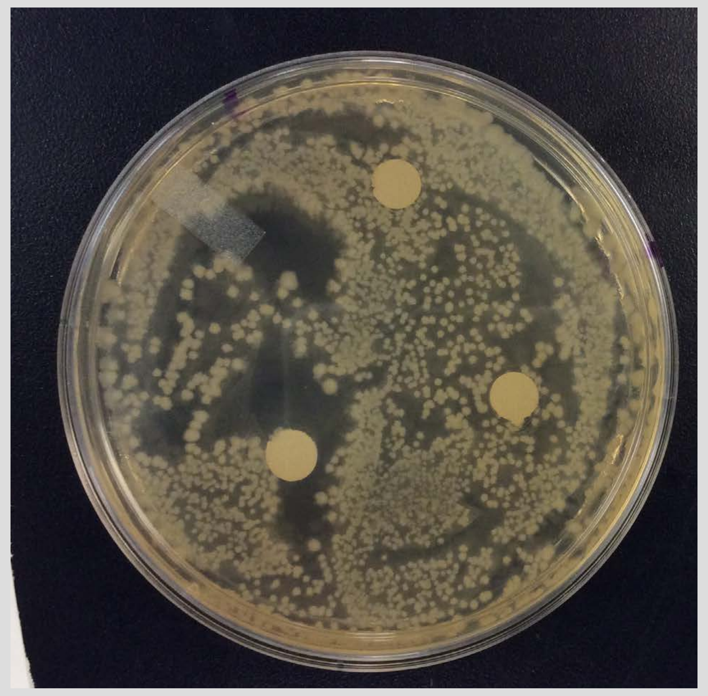
Kirby-Bauer Disk Diffusion Method

RESULTS

Results of the antimicrobial assays were produced after 24 hours of incubation of the plates. They came in the form of zones of exclusion radii around the paper disks: larger radii signified greater antimicrobial properties of the corresponding plant, while smaller radii signified poorer antimicrobial properties. Zones of exclusion varied from 0mm for the negative control to 13mm for the positive control, showing that the experiment worked correctly, as it should have.


Coffeeberry Tree Bark
4mm

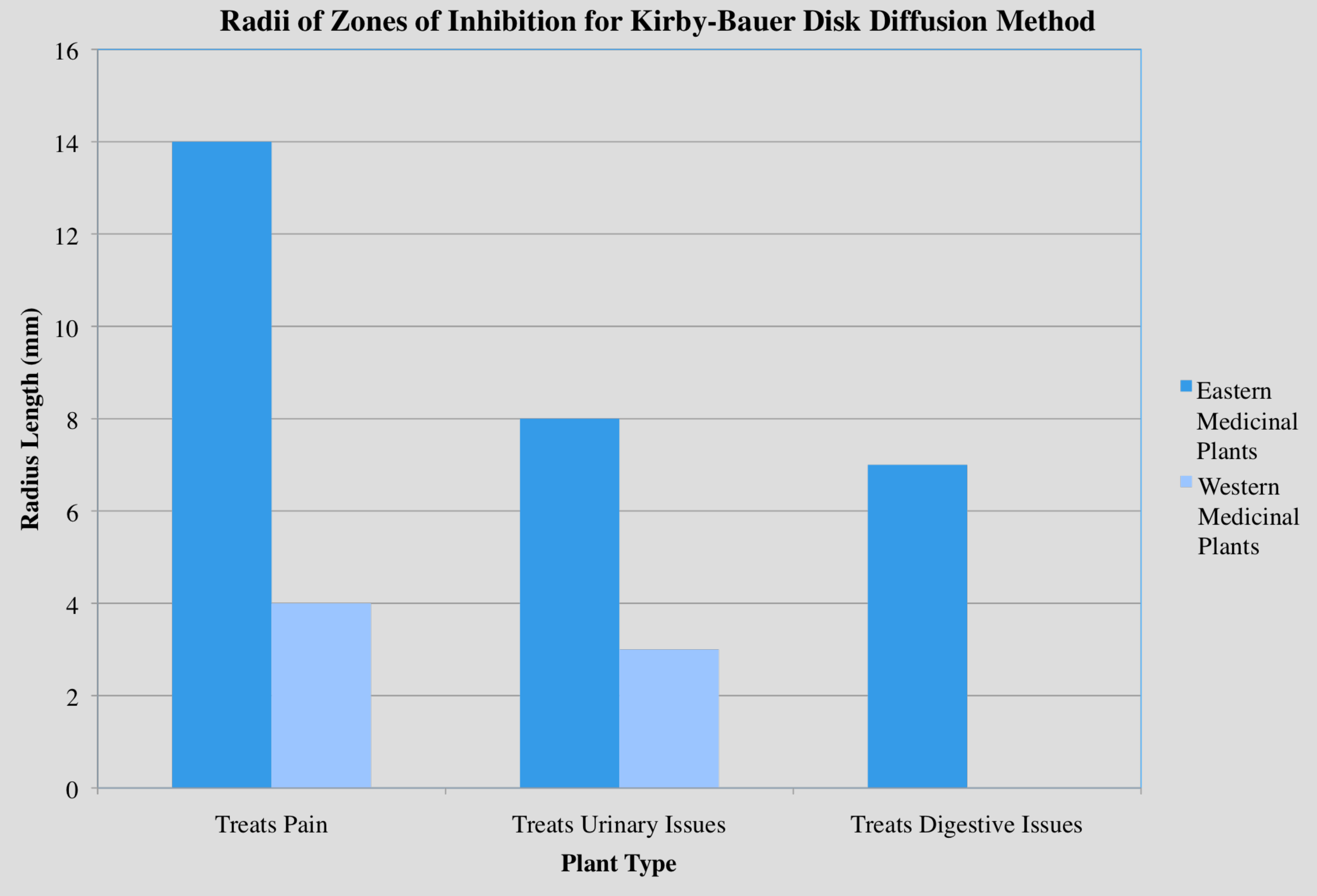

Manzanita Tree Bark
3mm


Redbud Tree Shoot Bark
0mm


Ampicillin (1g/1mL)
Positive Control
13mm


De-ionized Water
Negative Control
0mm

Below is a graph summarizing the results of the experiment. It compares the effectiveness in terms of zone of inhibition radii of the corresponding Eastern and Western medicinal plants according to the symptom they treat. The plants that treat pain are Coffeeberry tree bark (western) and Cinnamon bark (eastern). The plants that treat urinary issues are Manzanita tree bark (western) and Chinese yam (eastern). The plants that treat digestive issues are Western Redbud shot bark (western) and Chinese ginseng (eastern).

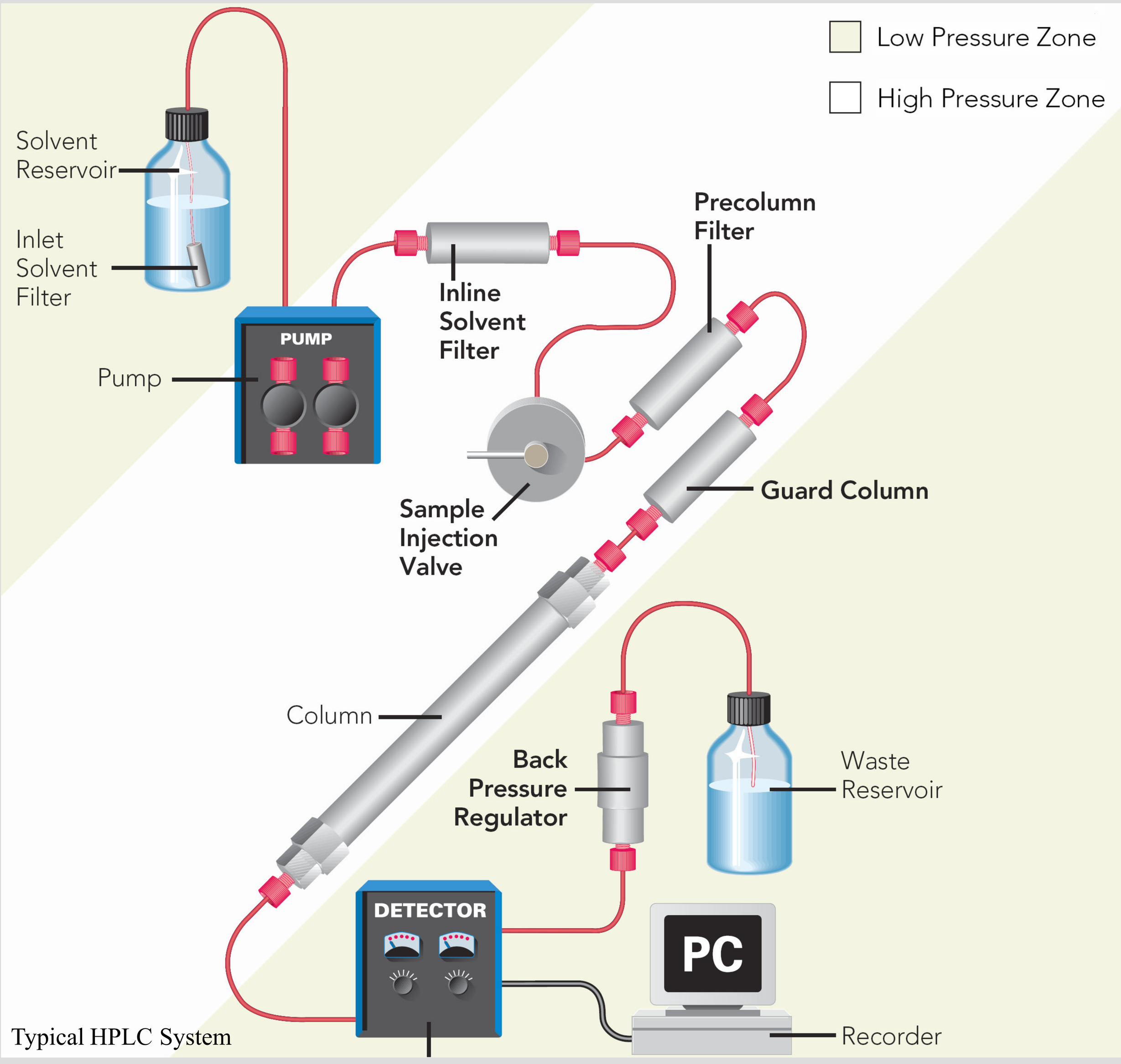


SUMMARY / CONCLUSIONS

The results of this experiment show several points clearly which are expressed by both the raw data and the graph. First, eastern medicinal plants are generally far more effective than Western medicinal plants. Their zone of exclusion radii were much larger than their corresponding western plants in every case, while one western plant proved to be entirely ineffective with a radius of 0mm. This is suggestive that there is a larger cause for this trend, possibly that the soil or other growing conditions in Asia and the eastern hemisphere are more favorable and provide better properties to plants that grow in them.
Additionally, medicinal plants that treat pain generally have more antimicrobial properties than those that treat urinary issues, which in turn have more antimicrobial properties than plants that treat digestive issues. This trend occurs in both eastern as well as western medicinal plants, and it suggests that bacteria are more involved in ailments that cause pain than they are in ailments of the digestive and urinary tracts. Because plants that treat pain are better at killing bacteria, they may also be effective in treating other symptoms that are caused by bacteria, such as food poisoning, meningitis, and gonorrhea.
Finally, the results suggest that plant components that are not tree bark are somewhat less effective at treating ailments than those that are. Overall, the plants with the greatest antimicrobial properties for both eastern and western medicines were tree barks, while the plants with the least antimicrobial properties for both eastern and western medicines were not. This is useful information because tree bark is one of the more accessible forms of plant that can be used for medicinal purposes, so medicines created from this type of plant are likely to be both cheaper and more effective.

FUTURE WORK

The next step of this experiment is to identify the specific metabolites contained within each plant that give them antimicrobial and other favorable properties. This process would require the use of a HPLC, or high-performance liquid chromatography machine. With this data, more information could be further attained in order to know exactly what compounds are providing the plants with favorable, health-improving qualities. These compounds could potentially be isolated and used to create or improve medicines that would treat these same symptoms, and possibly even more.



Typical HPLC System

ACKNOWLEDGEMENTS / REFERENCES

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Image Credits
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