



# SCREENING OF MEDICINAL PLANT EXTRACTS FOR ANTIBACTERIAL ACTIVITY AGAINST CLOSTRIDIUM DIFFICILE INFECTION



CONACYT

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## 1

### INTRODUCTION

The prolonged use of antibiotics is closely related to the infection by Clostridium difficile, Mexico is a country with incredible biodiversity of plants, and an alternative treatment is natural compounds with antibacterial properties. The main objective of this study was to analyze the effect of medicinal plants with an antibacterial action against toxigenic clinical strains of Clostridium difficile.

## 2

### METHODS

The products were selected for previous plant-based research that had evidence of use by rural communities in Mexico throughout history. A total of 24 different extracts were tested against toxigenic strains of Clostridium difficile against 6 clinical C. difficile isolates, and reference strain 027. The antibacterial activity, synergy, and antagonism of the extracts and the synergy between the extracts and antibiotics were evaluated using the Kirby Bauer disk diffusion method. (Hudzicki, 2016). The diameters of the zones of inhibition were measured in mm. Each test was performed in triplicate. The results were subjected to a one-way Anova analysis of variance, and mean comparisons were made by Tukey's multiple range test using the ASTATSA software (<https://astatsta.com>).

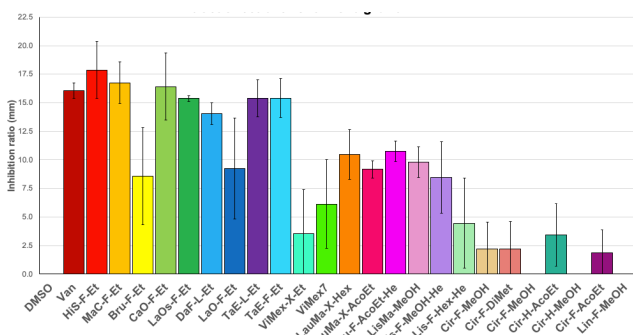


Figure 1. Extract effect over C. difficile growth

Extract	Inhibition ratio (mm)	
	Mean	Standar deviation
HiS-F-Et	17.58	2.35
MaC-F-Et	16.75	1.80
CaO-F-Et	16.42	2.88
LaOs-F-Et	15.38	0.26
TaE-L-Et	16.04	1.37
TaE-F-Et	15.42	1.71

Table 1. Inhibition zone (mm) of bacterial growth by plant extracts

## 3

### RESULTS

We identified six plant extracts that have an effective action (inhibition index > 15 mm) against all strains of C. difficile (Figure 1):

- HiS-F-Et (Roselle flower)
- MaC-F-Et (Chamomile flower)
- CaO -F-Et (Calendula flower)
- LaOs-F-Et (Lavender flower)
- TaE-L-Et (Marigold leaves)
- TaE-F-Et (Marigold flower)

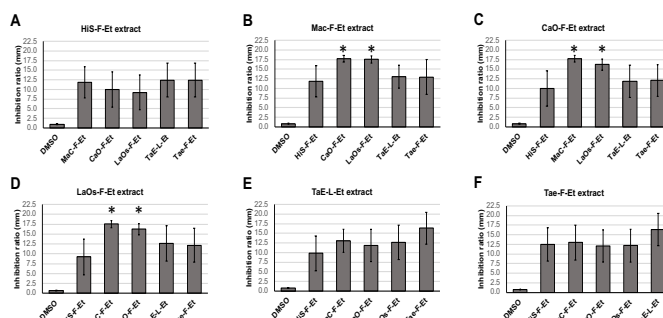


Figure 2. Synergic effect between plant extracts. A. Synergy with HiS-F-Et extract. B. Synergy with Mac-F-Et extract. C. Synergy with CaO-F-Et extract. D. Synergy with LaOs-F-Et extract. E. Synergy with TaE-L-Et extract. F. Synergy with TaE-F-Et extract. Mixture 1:1 of the extracts or extract:DMSO was analyzed by disc diffusion. Mean of three independent assays were represented in the graphs and statistical significance (p<0.05) were represented with an asterisk (\*).

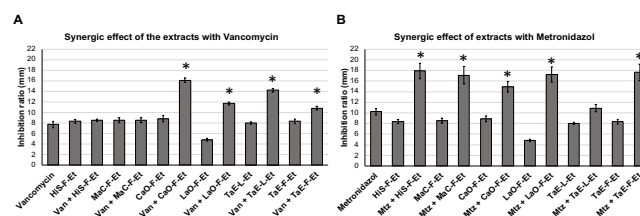


Figure 3. Synergic effect between the extracts and the antibiotics against CDI. A. Synergic effect of the extracts with Vancomycin. B. Synergic effect of the extracts with Metronidazol.

## 4

### CONCLUSIONS

Our results showed that vancomycin has synergy with Chamomile, Marigold and Cempasuchitl leaves; while metronidazole has synergy with Roselle, Chamomile, Lavender, Marigold, and Cempasuchitl flower.

By themselves, 6 of the 24 extracts show statistically significant inhibition of bacterial growth, while in the synergy between extracts 3 of the mixtures increase the activity of each extract up to double. Also, the extracts potentiate the effect of antibiotics.

## 5

### REFERENCES

Hudzicki, J. (2016). Kirby-Bauer Disk Diffusion Susceptibility Test Protocol Author Information. American Society For Microbiology, December 2009, 1–13. <https://www.asm.org/Protocols/Kirby-Bauer-Disk-Diffusion-Susceptibility-Test-Pro>