Antimicrobial Effect of the Chloroform Phase of *Praxelis clematidea*

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ABSTRACT

Medicinal plants constitute an arsenal of chemicals that could be exploited by human to prevent microbial invasion. *Praxelis clematidea* R.M. King & Robinson belongs to the family Asteraceae. Plants from this family have been extensively studied for the development of new drugs and insecticides. Based on this information, the chloroform phase of *Praxelis clematidea* was evaluated for antibacterial and antifungal activity. Six bacterial strains and six fungal strains were used in the study for activities. Microdilution method was used for antibacterial and antifungal assay of the chloroform phase. The results were also compared with the standard drug, Chloramphenicol (100 µg/mL) and Nystatin (100 UI/mL). The obtained results showed activity of the chloroform phase against *Candida* species, in particular against *Candida albicans*, which highlights the immense antifungal potential of this plant species.

Keywords: Chloroform phase, *Praxelis clematidea*, antimicrobial effect, *Candida* species

INTRODUCTION

The rise in antibiotic-resistant microorganism in recent years has led to an increasing search for new antibiotics. In general, it has been possible to observe an increase in resistance of pathogenic viruses, bacteria, fungi and protozoa against known drugs. To overcome the drawbacks of the current antimicrobial drugs and to obtain more efficacious drugs, an antimicrobial drug having a novel mode of action should be developed.

This increasing bacterial resistance is prompting resurgence in research of the antimicrobial role of herbs against resistant strains. A vast number of medicinal plants have been recognized as valuable resources of natural antimicrobial compounds.

Medicinal plants constitute an arsenal of chemicals that could be exploited by human to prevent microbial invasion. Secondary metabolites produced by plants constitute a major source of bioactive substances. The scientific interest in these metabolites has increased today with the search of new therapeutic agents from plant source, due to the increasing development of the resistance pattern of microorganisms to most currently used antimicrobial drugs.

Medicinal plant extracts offer considerable potential for the development of new agents effective against infections that are currently difficult to treat. Previous studies have shown that several substances such as peptides, unsaturated long chain aldehydes, essential oils and alkaloid constituents of plant extracts have potential therapeutic properties. Therefore, assessment of such plants remains an interesting and useful task to find new promising agents against bacterial infections. *Praxelis clematidea* R.M. King & Robinson belongs to the Eupatorieae tribe of the family Asteraceae, and consists of 2,400 species distributed in 170 genera. Plants from this family have been extensively studied for their chemical composition and biological activity and some have led to the development of new drugs and insecticides.

In phytochemical studies with ethanolic extract of *Praxelis clematidea* was isolated six flavonoids. This class is increasingly becoming an object of investigation, and many studies have isolated and identified flavonoids that possess antifungal, antiviral and antibacterial activities. In addition, various studies have demonstrated synergy between active flavonoids, and between flavonoids and conventional chemotherapeutic agents. Based on promising source of antimicrobial effects provided by species of the family Asteraceae, in particular those containing species flavonoid as secondary metabolites. The aim of the present study were to investigate the antimicrobial effects of Chloroform Phase of the aerial parts of *Praxelis clematidea* R.M. King & Robinson.

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MATERIALS AND METHODS
Preparation of plant extract
The aerial parts of Praxelis clematidea R.M. King & Robinson were collected in Lagoa do Paturi, a municipality of Santa Rita, in the state of Paraiba (Brazil), in May 2008. The identification of the botanical material was performed by Prof. Dr. Maria de Fatima Agra, Botany Sector, Laboratory of Pharmaceutical Technology/UFPB “Professor Delby Fernandes de Medeiros”. Exsiccates of the plant are deposited in the Prof. Lauro Pires Xavier (JPB) Herbarium, Paraiba Federal University, under the code M. F. Agra et al. 6894 (JPB). Maia et al (2010) describe the method of obtaining the chloroform phase\(^{15}\).

Bacterial and fungal strains
For antibacterial activity assays, were selected 6 strains of bacteria (Staphylococcus aureus - ATCC 13150, Staphylococcus aureus - ATCC 25923, Pseudomonas aeruginosa - P03, Pseudomonas aeruginos - ATCC 25853, Escherichia coli - ATCC 25922 and Escherichia coli - 5) and for antifungal activity assays, were selected 6 strains of fungi (Candida albicans – ATCC 90028, Candida albicans – LM 109, Candida tropicalis - ATCC 13803, Candida tropicalis – LMP 20, Candida krusei – LM 13 and Candida krusei – LM 08). All the microorganism strains were obtained from the Laboratory of Mycology collection. Bacteria and fungi were kept on Nutrient Agar (NA) slants at 4 °C. Inocula were obtained from overnight cultures grown on NA slants at 37 °C and diluted in sterile saline solution (NaCl 0.85% w/v) to provide a final concentration of approximately 10\(^{6}\) count forming unit per mL (cfu.mL\(^{-1}\)) adjusted according to the turbidity of 0.5 McFarland scale tube.

Antimicrobial and antifungal assay
The microplate bioassay was used to determine the minimum inhibitory concentration (MIC) of chloroform phase\(^{18,19}\). The antibacterial and antifungal activity was detected using the colorimetric method by adding 200 µL of resazurin staining (0.1 g.100 mL\(^{-1}\)) aqueous solution in each well at the end of the incubation period. MIC was defined as the lowest chloroform phase concentration able to inhibit the bacterial or fungi growth as indicated by resazurin staining (dead cells were not able to change the staining color by visual observation – blue to red)\(^{20}\). All experiments were carried out at least twice with consistent results.

RESULTS
The results for antibacterial activity of the chloroform phase of *Praxelis clematidea* (CFPC) are show in Table 1. Moreover, the results for antifungal activity of the CFPC are show in Table 2. The activity, in both cases, was measured in terms of presence of microorganism growth. Results obtained from the *in vitro* antibacterial assay showed that the CFPC show no antibacterial activity against either gram (+) or gram (-) bacteria. However, results obtained from the *in vitro* antifungal assay showed that the CFPC show promising antifungal activity against *Candida albicans* (ATCC 90028) with MIC of 32 µg/mL, and low antifungal activity against *Candida krusei* (LM 08) with MIC of 1024 µg/mL.

**DISCUSSION**

Resistance to available antibiotics is increasing at a very alarming stage globally. Efforts are urgently needed to replace current available antibiotics. In this context, the antibacterial activity of plants is continuously attracting global attention.

Many plants have been used because of their antimicrobial traits, which are due to compounds synthesized in the secondary metabolism of the plant. These products are known by their active substances, for example, the phenolic compounds that are part of the essential oils, as well as in flavonoids. The results obtained from the chloroform phase showed a significant and important antifungal effect against *Candida albicans*. Conventionally, treatment for candidiasis is usually done with the topical and oral administration of antifungal azole and polyene, but has been making frequent presence of such resistance to these microorganisms drugs because their inappropriate use. Furthermore, these drugs can cause toxic effects and considerable side, the decreases patient acceptance. Thus, the use of medicinal plants as medicine traditional proves to be quite attractive as an alternative therapy, requiring studies science on the subject, which are still insufficient.

In addition, the different behavior observed between strains of the same species could be justified by the existence of genetic variability among different strains. This antifungal activity against *Candida albicans* of CFPC has been observed in other studies with extracts of plant species of the family Asteraceae and is showed next to the results obtained with the ethanol extract of the same plant.

**CONCLUSION**

Based on these results it can be stated that the CFPC has an important antifungal activity against Candida species, which highlights the need for further studies with other fungal species to investigate the immense therapeutic potential of this plant species and with its isolated secondary metabolites.

**REFERENCES**


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