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Cymbopogon sp. from ethnobotany to antimicrobial: an integrative review

Cymbopogon sp. da etnobotânica ao antimicrobiano: uma revisão integrativa

Cymbopogon sp. de la etnobotánica a los antimicrobianos: una revisión integrative

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Abstract
Considering the broad potential of the genus *Cymbopogon*, here we present a systematic literature survey on it's antimicrobial potential. This is a review, articles from the Scielo and PubMed platforms. The articles surveyed were published between 2015 and 2020, with the theme of microbiology, including ethnobotanical studies, literature reviews, *in vitro*, *in vivo*, reports of clinical trials. Works outside the selected period, duplicate articles, and those only reporting infections of plants by the microorganism were excluded. 98 studies were selected, 74% found in PubMed and 26% in Scielo. Of this total, 21% were ethnobotanical/ethnopharmacology or literature reviews, most of them reporting the use of infusions of the species *C. citratus* (DC.) Stapf. Moreover, in 57% of the studies, survey respondents did not report antimicrobial use. In relation to 79% of the experimental studies, it was observed that 77% reported total inhibition of microbial growth, 3% indicated moderate growth inhibition, 4% low growth inhibition and 5% reported no inhibition. Among the microbial species analyzed were *Escherichia coli*, *Pseudomonas aeruginosa*, *Borrelia burgdorferi*, *Candida albicans*, *Salmonella enterica* and *Saccharomyces cerevisiae*. In addition, 5% of the articles reported antiviral activity, 5% parasitic control, 1% preventive action against contamination by mesophilic microorganisms. Although the population is not aware of the antimicrobial activity of *Cymbopogon* sp., studies have demonstrated its antimicrobial potential, thus the extracts of this genus can be an alternative for use in folk medicine as well as a source of new drugs with antimicrobial action.
Keywords: Plants; Medicinal; Phytotherapy; Ethnopharmacology; Poaceae; Lemongrass; Microbiology.

Resumo
Considerando o amplo potencial do gênero *Cymbopogon*, apresentamos aqui um levantamento sistemático da literatura sobre seu potencial antimicrobiano. Trata-se de uma revisão, artigos das plataformas Scielo e PubMed. Os artigos pesquisados foram publicados entre 2015 e 2020, com a temática da microbiologia, incluindo estudos etnobotânicos, revisões de literatura, *in vitro*, *in vivo*, relatórios de ensaios clínicos. Foram excluídos trabalhos fora do período selecionado, artigos duplicados e aqueles que relatavam apenas infecções de plantas pelo microrganismo. Foram selecionados 98 estudos, 74% encontrados no PubMed e 26% no Scielo. Desse total, 21% eram

etnobotânicos/etnofarmacológicos ou revisões de literatura, a maioria relatando o uso de infusões da espécie *C. citratus* (DC.) Stapf. Além disso, em 57% dos estudos, os entrevistados não relataram uso de antimicrobianos. Em relação a 79% dos estudos experimentais, observou-se que 77% relataram inibição total do crescimento microbiano, 3% indicaram inibição moderada do crescimento, 4% baixa inibição do crescimento e 5% não relataram inibição. Entre as espécies microbianas analisadas estavam *Escherichia coli, Pseudomonas aeruginosa, Borrelia burgdorferi, Candida albicans, Salmonella enterica* e *Saccharomyces cerevisiae*. Além disso, 5% dos artigos relataram atividade antiviral, 5% controle parasitário, 1% ação preventiva contra contaminação por microrganismos mesófilos. Embora a população desconheça a atividade antimicrobiana de *Cymbopogon* sp., estudos têm demonstrado seu potencial antimicrobiano, assim os extratos deste gênero podem ser uma alternativa para uso na medicina popular bem como fonte de novos fármacos com ação antimicrobiana.
Palavras-chave: Plantas; Medicinais; Fitoterapia; Etnofarmacologia; Poaceae; Capim-limão; Microbiologia.

Resumen
Considerando el amplio potencial del género *Cymbopogon*, aquí presentamos un estudio sistemático de literatura sobre su potencial antimicrobiano. Es una revisión de artículos de las plataformas Scielo y PubMed. Los artículos investigados fueron publicados entre 2015 y 2020, con el tema de microbiología, incluyendo estudios etnobotánicos, revisiones de literatura, *in vitro, in vivo*, informes de ensayos clínicos. Se excluyeron trabajos fuera del período seleccionado, artículos duplicados y aquellos que solo reportaron infecciones de plantas por microorganismos. Se seleccionaron 98 estudios, 74% encontrados en PubMed y 26% en Scielo. De este total, 21% fueron revisiones de literatura o etnobotánica/etnofarmacología, la mayoría reportando uso de infusiones de la especie *C. citratus* (DC.) Stapf. Además, en 57% de los estudios, los encuestados no informaron uso de antimicrobianos. Con relación al 79% de los estudios experimentales, se observó que 77% reportó inhibición total del crecimiento microbiano, 3% indicó inhibición moderada del crecimiento, 4% baja inhibición del crecimiento y 5% no informó inhibición. Entre las especies microbianas analizadas se encuentran *Escherichia coli, Pseudomonas aeruginosa, Borrelia burgdorferi, Candida albicans, Salmonella enterica y Saccharomyces cerevisiae*. Además, 5% de los artículos reportaron actividad antiviral, 5% control parasitario, 1% acción preventiva contra contaminación por microorganismos mesófilos. Aunque la población no conoce la actividad antimicrobiana de *Cymbopogon* sp., estudios han demostrado su potencial antimicrobiano, por que los extractos de este género pueden ser una alternativa para su uso en la medicina popular así como fuente de nuevos fármacos con acción antimicrobiana.
Palabras clave: Plantas medicinales; Fitoterapia; Etnofarmacología; Poaceae; Lemongrass; Microbiología.

1. Introduction

The use of medicinal herbs preparations to treat malignancies does not have a well-established origin, but reports confirm their relevance for the development of medicine and societies. Popular knowledge is the main source of information about this practice, and this knowledge is passed down within generations of a family or community (Rodrigues et al., 2017).

Due to the use of medicinal plants in many cultures, this use has been the subject of scientific knowledge and later integrated into public health policies, especially in primary care. This occurs through the adoption of integrative and complementary practices (ICPs), including the implementation of projects such as *Farmácias Vivas* (“Living Pharmacies”) in basic health units in Brazil (Brasil, 2016; Brasil, 2018; Ferreira et al., 2020).

The plants used in folk medicine need to be investigated for pharmacological activity and toxicity to ensure the safety of patients, because contrary to popular belief that natural substances will cause no harm, some plants used erroneously can be highly dangerous (Goulart et al., 2020).

Medicinal plants have several activities, such as regulation of the central nervous system, alleviation of inflammation, and maintenance of the biochemical or hormonal characteristics of the body. In addition to these uses, some plants have promising antimicrobial activity or precursor molecules of this class of drug (De Castro & Figueiredo, 2020).

Microorganisms are composed of parasites (arthropods, helminths, and protozoa), viruses, bacteria and fungi, with the ability to infect or parasitize humans and animals. Some can trigger a pathophysiological process that is treatable with a specific pharmacotherapy. However, due to the high genetic mutability of some of these microbes, a constant search for new antimicrobials is required, usually originated from molecular changes in existing drugs or secondary metabolites of plants, which are responsible for the antimicrobial action of these herbs (Oliveira et al., 2019; Sahal et al., 2020).

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Some botanical genera have been widely investigated regarding their antimicrobial activity. This is the case of *Cymbopogon* Spreng., a herb belonging to the angiosperm group, being classified as a monocotyledon of the Poaceae family (Gramineae). It is an aromatic species characterized by having long leaves with parallelelineal leaf rib, and rarely present flowers (Lorenze & Matos; 2008; Nguyen et al., 2019). Considering the broad potential of the genus *Cymbopogon*, here we present a systematic survey of the literature on its antimicrobial potential.

2. Methodology

This is a cross-sectional integrative review of the literature, a search for data available on the Scielo (https://www.scielo.org) and PubMed (https://.pubmed.ncbi.nlm.ni.gov) platforms for full articles published equating as contained information, following the processes and organized by the PRISMA protocol, following the investigative question: "*Cymbopogon* sp. does it have antimicrobial potential? (Marconi & Lakatos, 2017).

The term *Cymbopogon* was used as descriptor, adopting as inclusion criteria papers published from January 2015 to January 2020 in Portuguese, Spanish or English, with thematic area of literature review, ethnobotany, ethnopharmacology or microbiology involving *in silica, in vitro, in vivo* and clinical trials. Works outside the selected period, duplicate articles, and those where the microorganism infected only plants were excluded. Subsequently, these data were tabulated and quantified, with subsequent identification of the percentages of each variable.

3. Results and Discussion

In the study, 431 publications were analyzed, with the selection of 98 scientific works according to the inclusion and exclusion criteria. Among the selected articles, 74% (73/98) were found on the PubMed platform and 26% (25/98) in Scielo. This difference in the number of publication was possibly due to the scope of each of the platforms, PubMed has a larger number of indexed journals, from countries throughout the world, while Scielo has more restrictions on publications and its target audience is mainly from Latin American countries is restricted mainly to articles published by researchers from Latin American countries. The average annual publication was approximately 16 articles involving this theme, and the year with the highest number of publications was 2015 when considering both platforms, and 2018 when considering only PubMed.

3.1 Ethnobotany / Reviews

Among the variety of studies, 21% (21/98) were ethnobotanical, ethnopharmacological or literature reviews. After a detailed analysis of these studies, it was found that 57% (12/21) were surveys in which community respondents did not report the use of *Cymbopogon* species to treat any infection or symptom, while in 43% (9/21) of the articles of this type, the use of plants of this genus was reported to treat pathologies caused by microorganisms, such as influenza, pneumonia, fungal diseases and parasite infections. This showed that despite the wide popular knowledge about the *Cymbopogon* genus, some communities are still not aware of its antimicrobial potential. However, studies such as Cunha et al. (2020), Sahal et al. (2020) and Ahmad and Viljoen (2015) have demonstrated this activity against bacteria, fungi and viruses.

The main species cited was *Cymbopogon citratus* (DC.) Stapf, with 63% (19/30), considering that some studies cited more than one specimen of *Cymbopogon*. Regarding the form of preparation/extraction indicated in the studies, infusion appeared in 71% (15/21), while 29% (6/21) reported other forms of preparation (Table 1). Analysis of ethnobotanical, ethnopharmacological and literature review articles revealed that most of the studies were conducted in Africa or Brazil.

Table 1 - Publications of ethnobotanical nature, ethnopharmacological and literature review, being observed biological activity, etiological agent, report of antimicrobial activity, popular name and species.

Study	Biological activity	Etiological agent	Activity	Popular name	Species	Reference
Etno	Antimycobacteria	*Mycobacterium tuberculosis*	NAR	Lemongrass	*C. giganteus*	Nguta et al. 2015
Etno	Fever/ Cramps/ Cold/Cough	Without	NAR	Lemongrass	*C. citratus*	Clement et al., 2015; Kpodar etal., 2016
Etno	Soothing/ Pneumonia/ Digestive Disorders/ Spasmolytic / Flu	Without	AR	Capim-Cidreira	*C. citratus*	Paredes et al., 2015; Bieski et al., 2015; Intriago et al., 2015; Miguéis et al., 2019; Caetano et al., 2015
Rev.	Antifungal/ Antimalarial/ Anti-inflammatory/ Antimutagenic/ Anxiolytic/Antioxidant/ Hypoglycemia/ Hyperlipidemia	*Escherichia coli/ Proteus vulgaris/ Klebsiella pneumoniae/ Aspergillus* spp/ *Haemophilus influenza/ Pseudomonas aeruginosa/ Streptococcus aureus/ Streptococcus pyogenes/ Staphylococcus aureus/ Candida albicans/ Malassezia pathogenic*	AR	Lemongrass	*C. citratus*	Suroowan; Mahomoodally, 2016; Ekpenyong; Akpan; Nyoh, 2015; Donato et al., 2020
Rev.	Antifungal/ Hepatoprotective/ Antioxidant/ Antiprotozoario/ Antibacterial	Without	AR	Lemongrass	*C. parkeri/ C. olivieri/ C. citratus/ C. ambiguous/ C. flexuosus/ C. pendulus / C. proximus/ C. densiflorus*	Avoseh et al., 2015
Etno	Flu/ Stress Relief/ Inflammation/ Fever/High Cholesterol/High Blood Pressure/Indigestion	Without	AR	Capim-Santo	*C. citratus*	De Santana; Voeks; Funch, 2016
Etno	Repellent	Without	NAR	Kyayisubi	*C. citratus*	Baana; Angwech; Malinga, 2018
Etno	Antiparasitologic	Without	AR	Lemongrass	*C. citratus*	Odoh et al., 2018
Rev.	Antifungal/ Hepatoprotective/ Antioxidant/ Antiprotozoario/ Antibacterial	Without	AR	Citronela	*C. citratos/ C. winterianus*	Santos et al., 2019
Etno	Analgesia	Without	NAR	Lemongrass / Verbena	*C. citratus*	Mota et al. 2020
Etno	Colon reactivity	Without	NAR	-	*C. schoenanthus*	Djemam et al., 2020
Etno	Comforting	Without	NAR	Lemongrass	*C. citratus*	Santos; Silveira; Gomes, 2015
Rev./ Etno	Central Nervous System Disorders	Without	NAR	Capim-Cidreira/ Capim-Cidrão	*C. citratus*	Amoateng et al., 2018; Gross et al., 2019.

Rev: Review; Etno: ethnobotanical/ethnopharmacology; NAR: No Activity Report; AR: Activity Report. Source: Own authorship (2022)

C. citratus, popularly known as lemongrass or capim-santo, is one of the most common species of this genus, spread worldwide due to its adaptation to tropical conditions. It is one of the most known among the plants of the genus *Cymbopogon*, and is also widely used in folk medicine to treat gastrointestinal and central nervous system disorders (anxiolytics). However, some reports indicate its use to treat other symptoms or diseases, often correlated with infections such as the fever, pain and cough related to flu, malaria and pneumonia. Regarding the extraction mode, infusion to make tea was the most widely disseminated in communities (Ekpenyong, Akpan & Nyoh, 2015; Baana, Angwech & Malinga, 2016; Donato et al., 2018).

3.2 Experimental works

In 79% (77/98) most of the articles were experimental studies in which *in sílico, in vitro, in vivo* or clinical trials were performed to test the antimicrobial action of the genus *Cymbopogon*. Considering the resistance to antimicrobial therapies, a worldwide problem of complex epidemiology, these studies reported alternative therapies or sources of new molecules for the treatment of infections in humans and animals, as well as food safety (Pardo et al., 2018; Liu et al., 2019).

In the experimental studies, the use of eight different species of *Cymbopogon* was observed, while three studies did not report the species used. The species *C. citratus* was most cited, as well as in other types of works, followed by *C. nardus* (L.) Rendle, *C. flexuosus* (Nees) Will. Watson, *C. martini* (Roxb.) Will. Watson, *C. winterianus, C. schoenanthus* (L.) Spreng., *C. densiflorus* (Steud.) Stapf and *C. giganteus* Chiov. The studies of *C. citratus* reported variation in the sensitivity profile, as was the case of the species *C. nardus* and *C. martini*.

In addition, 73% (58/79) of the extracts/preparations used were essential oils (EOs), 11% (9/79) involved nanoparticles, 5% (4/79) were infusion, 8% (6/79) had other preparations and 3% (2/79) did not report the type of preparation.

Among these species reported in the studies, a variety of compounds were investigated. Citronellal, Myrcene, Citral and their chemical isomers stood out, as was the case of geraniol, a compound directly related to the antimicrobial potential of *Cymbopogon* (Trindade et al., 2015; Adukwu et al., 2016; Wu et al., 2019). Pontes et al. (2018) presumed that these compounds pass through the cell wall causing depolarization of the microorganism's plasma membrane (Miguéis et al., 2019).

This variation in the sensitivity profile may be related to seasonality, circadian cycle, soil and stress, generating changes in the synthesis of plant metabolites, which consequently can trigger different biological activities. Another factor influencing the sensitivity of microorganisms to these herbs was the mode of extraction/preparation. The main preparations found were essential oils followed by nanoparticles. The activity of these substances is already well elucidated in the literature, indicating antimicrobial potential. However, extracts that use solvents can present the active metabolite in a smaller amount, due to the dilution of these compounds in the solvent (De Santana et al., 2015; Seibert et al., 2019).

In the analyzed studies, tests were reported in strains of 52 genera and 98 distinct species. Of this total number of species, 63% (60/96) were bacteria, 29% (28/96) fungi (filamentous or yeast), 4% (4/96) viruses and 4% (4/96 parasites (arthropods, helminths and protozoa).

In the analysis of the results of articles that evaluated antifungal and antibacterial action, 77% (60/78) reported total inhibition of growth, 3% (2/78) moderate inhibition, 4% (3/78) low growth inhibition and 5% (4/78) no microbial inhibition. Some of these were strains of *Escherichia coli* (with antimicrobial resistance), *Pseudomonas aeruginosa, Borrelia burgdorferi, Candida albicans* (with antimicrobial resistance), *Salmonella enterica* subsp. *enterica* serovar Typhimurium and *Saccharomyces cerevisiae*. In addition, 5% (4/78) reported antiviral activity, 5% (4/78) parasitic control and 1% (1/78) preventive action against contamination by mesophilic microorganisms.

Regarding the evaluation of plant extracts of *Cymbopogon* species, a variety of tests were performed, the main ones being the microdilution or disk diffusion tests, to determine the Minimum Inhibitory Concentration (MIC), Minimum

Bactericidal Concentration (MBC), Minimum Concentration of Biofilm Eradication, Fractional Inhibitory Concentration Index (FICI), and exposure with observation and cell culture.

The broth microdilution MIC test stood out in this review as the most used method, probably because it is a method recommended by the Clinical and Laboratory Standards Institute (CLSI) to determine drug inhibition concentrations against anaerobic bacteria growth, yeasts and filamentous fungi. This method was adapted according to the tested substances, for example involving concentration of the tested substances or their solubilization process. In relation to solubilization of the test compound, it was observed that in the case of essential oils, due to their apolar nature, a substance is required to solubilize them in the culture medium, such as Tween 20 or 80 and Dimethyl sulfoxide (DMSO). These substances are used in the lowest practical concentrations due to toxicity (Balouiri et al., 2016; Costa et al., 2019).

The disk diffusion method was reported to have good applicability due to its ease of conduction and low cost. However, it is not as precise as microdilution, because it depends on the diffusion of the bioactive substance in the medium and on the macro growth of the strains for the interpretation of the results, while microdilution test results can be read by absorbance and consequently produce more accurate inhibition results (Balouiri et al., 2016).

These methods are widely applied to extracts, especially essential oils derived as nanoparticles, in bioprospecting in the fight against various microorganisms. *In vitro* trials have been conducted in recent years to ascertain the antifungal potential against strains of *Candida albicans, Candida tropicalis, Candida krusei, Candida parapsilosis, Cryptococcus grubii, Penicillium corylophilum, Trichophyton rubrum, Trichophyton mentagrophytes, Microsporum gypseum* and *Microsporum canis*, pathogens that are sometimes neglected despite their clinical relevance. This is especially true of *Candida* sp., which has potential to develop biofilms and cause opportunistic infections in debilitated patients, especially in hospitals. In particular, it is the most common microorganism found in patients with oral cancer, whose incidence has been increasing significantly among young people and women, with 350,000 – 400,000 cases per year (De Toleto et al., 2016; Dias et al., 2017; Almeida et al., 2016; Gholizadeh et al., 2016; Khosravi et al., 2018; Da Silva Gündel et al., 2018; Córdoba et al., 2019; Ji et al., 2019; Silva et al., 2019; Sahal, et al., 2020).

Growth inhibition activity was also observed in gram-positive bacteria, such as *Staphylococcus epidermidis, Staphylococcus aureus, Enterococcus faecalis* and *Streptococcus mutans*, as well as against gram-negative bacteria, especially *E. coli, P. aeruginosa, Klebsiella pneumoniae, Acinetobacter baumannii* and *Proteus mirabilis*, showing a variation in relation to the results of the MIC and highlighting that non-fermenting bacteria, *P. aeruginosa* and *A. baumannii*, where less inhibition was seen (Scazzocchio et al., 2016; Chaftar et al., 2015; Leite et al., 2016; Souza et al., 2016; Rossi et al., 2017; De Silva et al., 2017; Luís et al., 2017; Bermúdez-Vásquez et al., 2019; Costa et al., 2019; Oliveira et al., 2019; Cunha et al., 2020).

In addition to studies showing inhibitory action against fungi and bacteria, some studies have investigated antiviral action of the essential oil of the species *C. citratus*, applied as a nanogel, against Herpes simplex viruses. The EOs of *citratus* and *nardus* were tested against the Mastadenovirus HAdV-5 and were found to have antiviral activity in both studies, with *C. citratus* also being effective in Murine norovirus 1 (MNV-1) and Dengue virus type 2 DENV-2 models. However, only a few studies have been published in recent years investigating the antiviral potential and action mechanism of these plants (Kim et al, 2017; Almeida et al., 2018; Chiamenti et al., 2019; Rosmalena et al., 2019).

Another group of microorganisms against which substances obtained from *Cymbopogon* sp. were studied was protozoa, namely *Plasmodium* sp., *Pediculus capitis, Giardia lamblia* and *Haemonchus contortus.* These studies showed the potential for prevention or control of growth of the populations of these parasites. However, only a few studies have been published, despite the potential of this plant (Macedo et al., 2015; Chukwuocha et al., 2016; Limoncu et al., 2017; Méabed, et al., 2018; Macedo et al., 2019).

There is strong global concern about multidrug resistance to conventional antibiotics. Thus, in 2019 the World Health Organization included antimicrobial resistance as one of the top ten global health threats (WHO, 2019). The findings of this review showed good effectiveness of *Cymbopogon* sp. against microorganisms, so it is important to conduct further studies to analyze the effectiveness of essential oils and their derivatives against drug-resistant microorganisms.

Singh, Fatima and Hameed (2016) reported citronella's potential against *C. albicans* in daily cleaning of dentures. Another use of the EOs is treatment of water and food, as indicated by Chaftar et al. (2015) and Boeira et al. (2018), who compared the effectiveness of different oils and extracts for control of crop pests and food rot.

Another way to use volatile oils of this botanical genus is with the addition of metallic nanoparticles, such as silver, or nanoemulsions, to enhance the biological activity of essential oils by preserving their constituents until contact with microorganisms, thus triggering more effective responses. This can be a strategy for use of natural products against resistant microorganisms (Bansod et al., 2015; Liakos et al., 2016; Rossi et al., 2017; Da Silva Gündel et al., 2018; Seibert et al., 2019; Cherian et al., 2020).

4. Conclusion

This integrative review of *Cymbopogon* allows us to conclude that the PubMed platform presented the largest number of publications during the period studied and in both platforms, the largest number of publications occurred in 2015. In the case of ethnobotany, ethnopharmacology and literature review articles, the interviewees were generally unaware of *Cymbopogon's* potential to treat infections.

The highest percentage of publications were experimental studies, of which more than half involving *Cymbopogon* species found antimicrobial potential against infectious agents, with *C. citratus* being the most studied species.

The great antimicrobial potential of medicinal plants of the genus *Cymbopogon* was revealed, thus recommending greater bioprospecting for the discovery of new drugs to combat microorganisms, with or without resistance to current pharmacotherapeutics. Therefore, further studies are needed to test secondary metabolites, their molecular variations and synergism with other drugs for application against multidrug-resistant microorganisms.

Cymbopogon derived EOs also have strong potential for the development of drugs with action against bacteria and fungi, as well as for food preservation. However, more studies still need to be carried out to evaluate the use of volatile oils against multidrug-resistant viruses, parasites and microorganisms due to their relevance in human and veterinary medicine. In addition, studies with nanoparticles and nanoemulsions have shown better microbiological performance compared to free EOs, thus being a possible source for the development of new pharmaceutical products. Furthermore, it is necessary to carry out characterization and isolation studies of EOs as well as toxicity studies to ensure repeatability and feasibility in the use of these substances.

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