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ETHNOBOTANICAL SURVEY OF MEDICINAL PLANTS USED FOR DISINFECTION OF PREMISES AND SURFACES IN THE FEZ-MEKNES CITIES (CENTRE OF MOROCCO)

S. Jaouhar ^{1,2}, K. Bekhti ^{*1}, J. El Oualidi ³, J. Bouzid ^{4,5} and A. El Ouali Lalami ^{1,2}

Laboratory of Microbial Biotechnology ¹, Faculty of Science and Technology, Sidi Mohammed Ben Abdellah University. PB 2202, Fez, Morocco.

Hassan First University of Settat ², Higher Institute of Health Sciences, Laboratory of Health Sciences and Technologies, 26000, Settat, Morocco.

Department of Botany and Plant Ecology ³, Herbarium National Rabat responsible, Scientific Institute. Avenue Ibn Battota PB 703 Agdal 10090 Rabat, Morocco.

Laboratory of Environment and Health ⁴, Faculty of Sciences Moulay Ismail University, PB 11201 Zitoune Avenue, Meknes, Morocco.

Higher Institute of Health Sciences ⁵, Hassan first University, Road of Casablanca km 3, 5 PB 555 Settat - Morocco.

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Correspondence to Author:

Bekhti Khadija

Laboratory of Microbial Biotechnology, Faculty of Science and Technology, Sidi Mohammed Ben Abdellah University. PB 2202, Fez, Morocco.

E-mail: bekhtii.khadija@gmail.com

ABSTRACT: The irrational and repetitive use of synthetic chemical disinfectants can contribute to the emergence of resistant strains and represent a risk to health and the environment. The aim of this ethnobotanical survey, a first innovative study, is exploring for plants with disinfecting power. An ethnobotanical study was conducted among herbalists in Fez and Meknes cities located in the center of Morocco. Information covered includes the vernacular and scientific name of plants, used part, mode of preparation, and administration. A total of 13 species belonging to 8 families has been described. Lamiaceae and Asteraceae (23%) were the most representative with three species every one, followed by the Myrtaceae (14%) with two species. The other families (Cupressaceae, Caryophyllaceae, Rosaceae, Rutaceae, and Brassicaceae), were represented with the same percentage (8%), a specie for each family. The most recommended species are *Lavandula* spp. (61%), *Origanum elongatum* (33%), *Artemisia herba-alba* (23%), *Rosmarinus officinalis* (17%), *Eucalyptus* spp (17%), *Atractylis gummifera* L. (16%), *Citrus limon* (8%), *Anastatica hierochuntica* (6 %), *Myrtus communis* L. and *Rosa damascena* (% for each), *Artemisia absinthium* and *Corrigiola telephiifolia* (2% for each) *Tetraclinis articulata* (3%). Leaves and flowers establish the most used parts. The incense was the most quoted mode of preparation. The disinfection by air contact was the recommended mode. This work would be of great interest to solve the problem of resistant strains and to develop effective biological disinfectants that respect the environment.

INTRODUCTION: The hospital environment is a place to risk; air and surfaces may be contaminated by different microorganisms from patients ¹.

The consequence of the contamination can lead to Healthcare-Associated Infections (HAI) ². So, adequate measures are necessary to reduce the risk of cross-contamination between the patients, the hospital environment, and the caring staff.

As a result, surfaces and medical equipment must be disinfected regularly once or several times a day ^{3, 4}. However, the effectiveness of disinfectants remains variable from application to another.

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Indeed, the massive and repetitive use of the same disinfectant with the same concentration can contribute to the emergence of resistant strains⁵. In addition, several have shown the emergence of bacterial resistance against different disinfectants⁶. On the other hand, these products represent a chemical risk for health professionals and the environment and are aggressive for medical devices^{7, 8, 9}. Indeed, various authors have shown that health care professionals routinely exposed to disinfection products used to control healthcare-associated infections^{10, 11}. For this purpose, several studies have shown that exposure to disinfectants generates serious health risks, especially respiratory disorders¹²⁻¹³, cutaneous infection, and cancers. It is reported that this risk has been associated with the use of quaternary ammoniums and bleach, products used for the disinfection of surfaces and medical instruments^{14, 15}.

Currently, scientists are looking for alternative products to synthetic chemicals such as herbal products. In fact, Aromatic and Medicinal Plants (AMPs) are often used as a source of bioactive compounds with pharmaceutical and agrochemical interest¹⁶. AMPs contain an amalgam of secondary metabolites, like terpenoids, alkaloids, flavonoids, and phenolic compounds whose antimicrobial activities have been demonstrated¹⁷. In the whole world, numerous scientific works relating to plants and their extracts (essential oils or aqueous extracts) have demonstrated their antifungal, antimicrobial, and antiviral properties¹⁸. In Morocco, different research works on AMP, and several ethnobotanical studies have been conducted. These studies have often focused on the use of AMP to treat diseases such as diabetes, high blood pressure, diseases of the digestive system or cutaneous infection^{19, 20, 21, 22}.

Thereby, AMP can also be used as herbal ecological bio-disinfectants for the hospital environment, especially as an alternative to synthetic chemical disinfectants used for air, surfaces, and materials. In this context, the aim of this first innovative study is to document and to explore disinfectant medicinal plants used by the Morocco population in the cities of Fes-Meknes (center of Morocco). To our knowledge, no study has focused on the use of AMP as bio-disinfectants. This work would be of great interest to health

professionals to solve the problem of resistant strains and for the pharmaceutical industry so as to develop an eco-friendly disinfectant.

MATERIALS AND METHODS:

Study Site: The city of Fez is the capital of the Fez-Meknes administrative region, **Fig. 1**. The prefecture of Fez covers an area of 332.1 km². It is located in Saïs Plain, halfway between north and south of the Kingdom of Morocco. The population of the prefecture is about 1 150 131 inhabitants; it represents 27% of the population of the region Fez-Meknes²³. The city of Meknes is located in North-western, Morocco. It is the former capital of the Meknès-Tafilalet administrative region and currently one of the two urban centers' of the Fez-Meknes region. It is located about 500 meters above sea level, on Saïs Plain between the Middle Atlas Mountains to the south and the Pre-Riff Hills to the north. The population of the prefecture of Meknes is about 835 695^{23, 24}. The climate of the studied area is a temperate type of the Mediterranean undergrowth, undergoing the continental influences during summer and winter. However, the geographical diversity of the region means each of its natural areas has particular climatic nuances²⁴. Note that, owing to its Mediterranean bioclimates and its geographical position, Morocco occupies the first place among the south of the Mediterranean because of its treasure-trove of endemic plants²⁵.

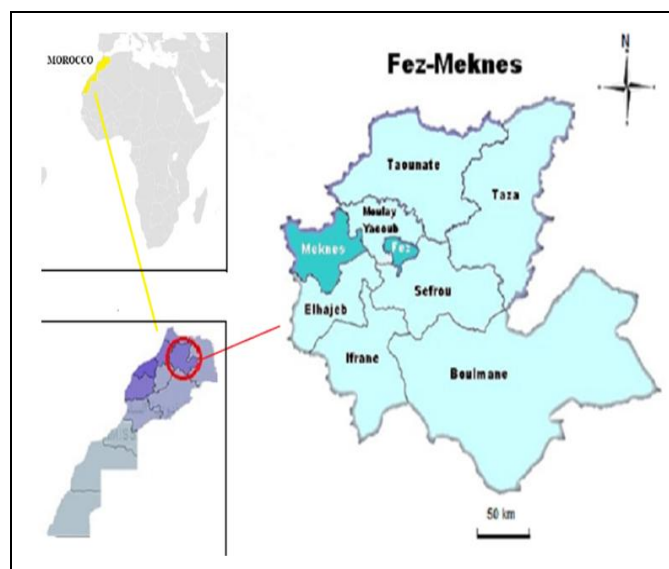


FIG. 1: MAP OF FEZ-MEKNES REGION

Source: High Commissioner for Planning, regional Direction of Fez-Meknes.

Conduct of the Study: In order to document local knowledge regarding the exploitation of medicinal and aromatic plants as a bio-disinfectant, an ethnobotanical survey was conducted over a period of 4 months (from October 2018 to January 2019). To carry out this survey, we went around the most popular areas of two main cities in the study area, Fez and Meknes (central Morocco). The number of people questioned was 63 herbalists. A preliminary interview was conducted with herbalists to get and to provide an overview of the purpose of the survey to obtain their consent to participate in the study. The conduct of the survey was based on semi-structured interviews with herbalists (pre-established interview guide) in the Moroccan dialect. The time spent with each herbalist varied from 30 to 45 min. The herbalists included in this study are not only sellers of plants, but they are also traditional healers.

The interviews were conducted with the help of a survey sheet which includes the following sections: i) the identity of the herbalist: age, sex, years of experience and level of study, ii) Plants that can be used for disinfection, iii) Plant information: Local name, Scientific name, Origin and Harvest period, iv) Directions for use: Part used disease, dose, preparation, and use methods, precautionary use, v) other uses of the plant. The identification of the scientific name of plants was realized at the scientific institute in Rabat by the Chief of Department of Botany and Plant Ecology who is responsible of Herbarium National of Rabat (The capital of Morocco).

Data Processing: The analysis of the data was performed by using the Excel software 2010 version and by the calculation of the frequency index (FI) according to the formula below:

$$FI = \Sigma N / n \times 100$$

Where ΣN = number of times the plant is cited, n = number of herbalists surveyed

RESULTS:

Information of Interviewed Herbalists: In the present survey, a total of 63 traditional herbalists and healers were interviewed for 45 min **Table 1**. All of the traditional herbalists were men more than 50 years ($n = 24$). The majority of herbalists had a Secondary level ($n = 22$); 18 had no education.

The average of years of experience was, the majority were between 11 and 20 years ($n = 24$). However, 22 of the herbalists had experienced between 21 and 30.

TABLE 1: CHARACTERISTICS OF RESPONDENTS (N=63)

Characteristic	Number
Gender	
• Male	63
• Female	0
Age between	
• 20–30 years	02
• 31–40 years	13
• 41–50 years	18
• 51–60 years	24
• 61-70 years	04
• >71 years	02
Education	
• No education level	18
• Primary level	10
• Secondary level	22
• Higher level	13
Years of experience between	
• 1–10 years	13
• 11–20 years	24
• 21–30 years	22
• 31–40 years	03
• 41–50 years	01

Inventory of Plants: In this study, 13 aromatic and medicinal plants were listed by herbalists as bio-disinfectants in **Table 2**. They belong to 8 botanical families with a predominance of the Lamiaceae family (23%) and Asteraceae (23%), followed by the Myrtaceae family (14%). The other families (Cupressaceae, Caryophyllaceae, Rosaceae, Rutaceae, and Brassicaceae), were represented with the same percentage (8%), a specie for each family **Fig. 2**.

According to the calculated frequency index (FI), the most recommended species are *Lavandula*. spp (FI = 61 %), *Origanum elongatum* (FI = 33 %), *Artemisia herba-alba* (FI = 23 %), *Rosmarinus officinalis* (FI = 17 %), *Eucalyptus*. spp (FI=17 %), *Atractylis gummifera* L. (FI=16 %), *Citrus limon* (FI=8 %), *Anastatica hierochuntica* (FI = 6 %), *Myrtus communis* L. and *Rosa Damascena* (FI = 5 % for each), *Artemisia absinthium* L. and *Corrigiola telephiifolia* (FI=2 % for each) *Tetraclinis articulata* (FI = 3 %) **Fig. 3**.

TABLE 2: THE ETHNOBOTANICAL INFORMATION OF 13 EFFECTIVE HERBS USED

Local name*	Nomenclature Scientifique	Families	Used part	Mode of preparation	Mode of administration
Zaater الزعتر	<i>Origanum elongatum</i>	Lamiaceae	Leaves	Decoction/Incense/Hydrolate	Contact with air or surface
Khzama الخزاما	<i>Lavandula. sp</i>	Lamiaceae	Leaves and Flowers	Decoction/Incense/ without preparation/ Aqueous Extraction	Contact with air or surface
Laraar العرعار	<i>Tetraclinis articulata</i>	Cupressaceae	Leaves	Incense	Contact with air
Rayhan الريحان	<i>Myrtus communis L.</i>	Myrtaceae	Leaves	Decoction/infusion	Contact with surface
Azir, Yazir أزير	<i>Rosmarinus officinalis L.</i>	Lamiaceae	Leaves/ All the aerial part	Decoction	Contact with air or surface
Chih الشيح	<i>Artemisia herba-alba</i>	Asteraceae	All the aerial part/the whole plant	Decoction/incense/ Aqueous Extraction	Contact with air
Chiba شيبا	<i>Artemisia absinthium L</i>	Asteraceae	Leaves/ All the aerial part	Decoction/Hydrolate	
Lcalitus لكاليتوس	<i>Eucalyptus sp</i>	Myrtaceae	Leaves	Decoction/Essential oil	
Dad ou الداد	<i>Atractylis gummifera</i>	Asteracea	Roots/Rhi zome	Incense	
Sarghina سرغينة	<i>Corrigiola telephifolia</i>	Caryophyllaceae	Roots/Rhizome	Incense	
Louard الورد	<i>Rosa Damascena</i>	Rosaceae	Flowers	Essential oil/ without preparation	Contact with air or surface
Lhamed الحامض	<i>Citrus limon</i>	Rutaceae	Fruit	Essential oil	Contact with air
Kaf Maryam كف مريم	<i>Anastatica hierochuntica</i>	Brassicaceae	All the aerial part	Hydrolate/ Aqueous Extraction	

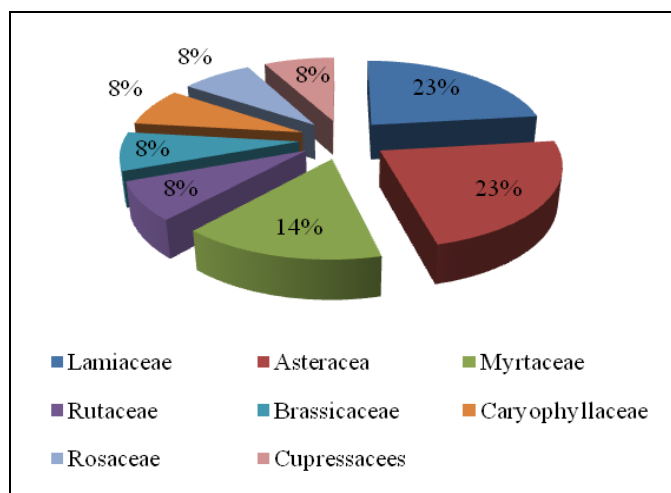


FIG. 2: PERCENTAGE OF DIFFERENT FAMILIES USED OF PLANTS

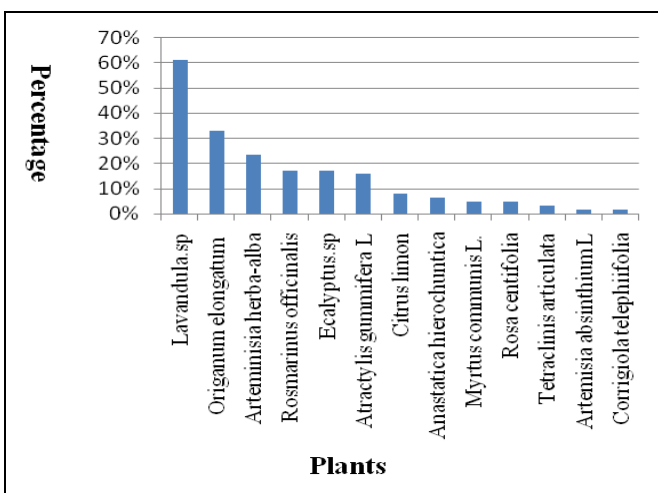


FIG. 3: FREQUENCY INDEX OF PLANTS

Used Part of Plants: In the study area, the most used parts of plants were leaves (37%), flowers (28%), leaves and flowers (14%) roots (7%), whole plant (6%), fruit (4%), the aerial part of the plant (3%) and rhizome (2%) **Fig. 4.**

Preparation and Administration Mode of Plants: In order to facilitate and make effective the administration of the active ingredients of plants, several methods of preparation are employed.

In the study area, most of the plants cited by herbalists, as disinfectants, should be prepared according to the following modes: incense (40.8%), decoction (28%), no preparation (14.4%), extract liquid (8%), essential oil (4%), infusion (3.2%) and hydrosol (1.6%) **Fig. 5.** The results of this study show that the disinfection of rooms and surfaces can be achieved by contact with air (68%) or by direct contact with surfaces (32%) **Fig. 6.**

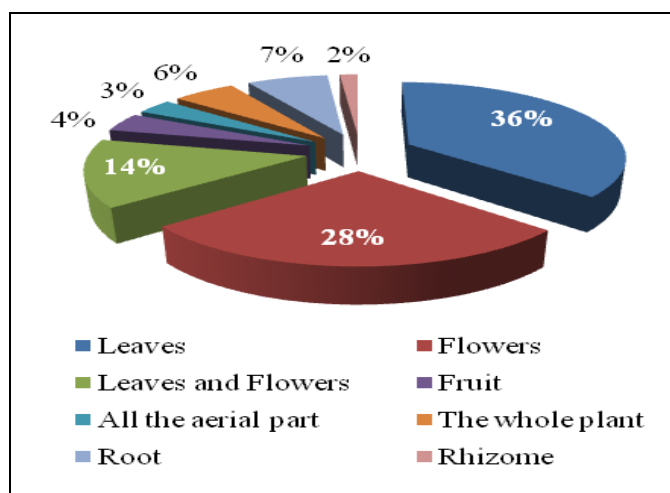


FIG. 4: PARTS OF PLANTS USED

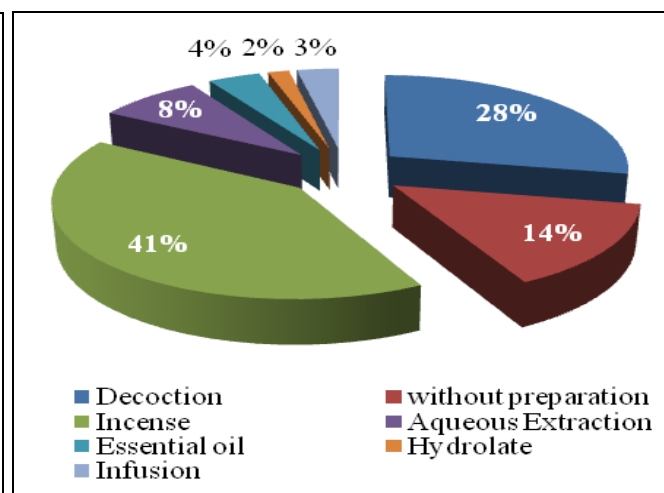


FIG. 5: MODE OF PREPARATION OF PLANTS

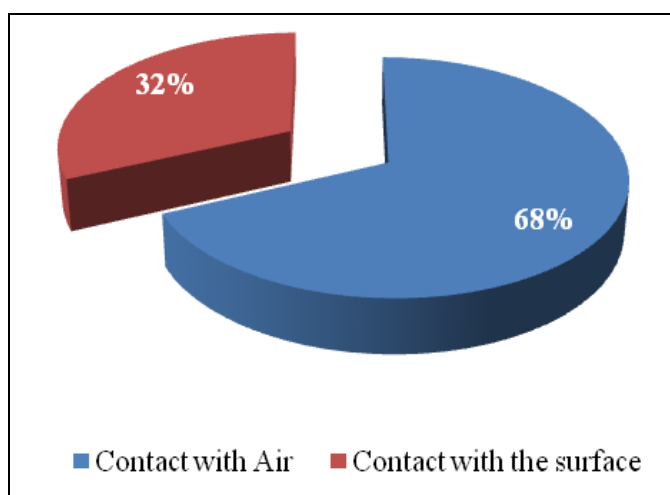


FIG. 6: MODE OF ADMINISTRATION OF PLANTS

DISCUSSION: Given the lack of studies on PAMs used as bio-disinfectants, we compared our results only with ethnobotanical surveys conducted on the "medical use of PAMs" aspect. In this study, 63 herbalists participated in the survey are male. This result could be explained by Moroccan culture, which does not encourage women to do this type of work. Thereby, our results are similar with ethnobotanical studies in Africa^{26, 27} and in Morocco²⁸. The average age was 46 years old, the majority were between 41 and 50 years old (n=24).

This result corroborates with the study of Bekalo *et al.*,²⁹ The majority of the herbalists had a Secondary level (n=21), while 18 of them are illiterate. Only 13 of the interviewed had a higher level. These results are not similar with those reported by Boufous *et al.*,³⁰ The predominance of Lamiaceae in Morocco and its high biodiversity has been reported in several studies^{31, 32}. Moreover, Lamiaceae and Asteraceae family's were also the

most reported in Moroccan ethno-pharmacological studies^{33, 34}. Indeed, the Lamiaceae are used by the Moroccan population for treatment of various pathologies^{36, 37}. The chemical composition of Lamiaceae is endowed with antimicrobial activity^{38, 39}. As noted above that we have not found ethnobotanical studies conducted, either nationally, about the use of medicinal plants as bio-disinfectants.

Most ethnobotanical studies have reported plant use as a treatment for parasitic diseases, respiratory and digestive infections, diabetes, heart disease and cancer^{40, 41, 42}. However, some plants inventoried in this study have been reported in the literature as a disinfectant. Indeed, in India and China eucalyptus is cited as a disinfectant plant^{43, 16}. This plant has shown a potential effect as a disinfectant of surfaces and hospital environments⁴⁴. Furthermore, many previous investigations have confirmed the antimicrobial activity of eucalyptus against various bacterial species, fungi and virus^{45, 46}. The good antibacterial activity of eucalyptus essential oil correlates with the amount of 1, 8-cineole, p-cymene, acinene or cryptone^{47, 46}.

Moreover, the antimicrobial activity of leaf extracts from 26 Eucalyptus species against Gram-positive bacteria (*Staphylococcus aureus*, *Bacillus cereus*, *Enterococcus faecalis*) has been proven, however, they have not shown any strong antibacterial activity against Gram-negative bacteria⁴⁸. The antibacterial and antifungal of lavender oil has been widely demonstrated^{49, 50}. The antimicrobial activity of this essential oil is probably triggered by its active compounds like Linalool acetate and β -

linalool⁵¹. The extract of *Rosmarinus officinalis* showed antibacterial effect on *Enterococcus faecalis* and capacity to disinfect guttapercha cones contaminated with it⁵². Antibacterial activity of essential oils of *Rosmarinus officinalis* against pathogenic bacteria isolated from hospital environment⁵³. The anti-microbial activity of *Artemisia herba-alba* against various bacterial species has been confirmed^{54, 55}. In addition, the essential oil of this plant exhibited much higher antibacterial activity against *Bacillus subtilis* ATCC and *Lactobacillus* sp⁵⁶.

About *Atractylis gummifera*, it has been cited in several ethnobotanical studies conducted in Morocco and it is used by the population as an antiseptic for skin^{57, 58}. However, this plant was found characterized by production of two highly toxic metabolites (attractyliside and carboxyattractyliside) responsible for frequent and fatal poisoning⁵⁹. The leaves' extract of *Origanum elongatum* showed also the most effective against the *Escherichia coli*⁶⁰. The other plants, that are rarely cited by the herbalists in the studied region (FI <10%), are recognized as well in several studies as antibacterial agents against nosocomial microorganisms. Indeed, the antimicrobial activity of topical formulation containing the essential oils of *Eugenia caryophyllata* and *Myrtus communis* were evaluated on selected skin disease, results indicated that the formulations have shown antimicrobial activity⁶¹.

The leaves' extract of *Myrtus communis* showed the most effective against the *Escherichia coli*⁶². The antibacterial activity of Citrus Limon against *Staphylococcus aureus*, *Enterococcus faecalis* and *Salmonella* spp has been demonstrated by Oikeh et al.,⁶³ Regarding *Rosa damascena*, Tofighi et al.,⁶⁴ have confirmed the antimicrobial activity of the extract of this plant against *Bacillus cereus*, *Staphylococcus epidermidis*, *Staphylococcus aureus* and *Pseudomonas aeruginosa*. In addition, essential oil of *R. damascena* showed antioxidant and antibacterial properties⁶⁵. All parts of *Anastatica hierochuntica* were reported to possess antimicrobial activity⁶⁶. Its freshly prepared decoction is commonly used as local disinfectant⁶⁶. The *Tetraclinis articulata* essential oil presents, *in-vitro*, a good activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia*

*coli*⁶⁷. The methanol extracts of *Corrigiola telephiifolia* revealed antibacterial activities against Gram-negative bacteria (*Pseudomonas aeruginosa*, *Klebsiella pneumonia*, *Escherichia coli*) and Gram-positive bacteria (*Staphylococcus aureus*, *Bacillus subtilis*, *Micrococcus luteus*)⁶⁸. All previously cited and studied bacteria were identified and found in the hospital environment of Fes city especially at surfaces and materials^{69, 70}. In the study area, the most used parts of plants were leaves. Our results are similar with several studies which clearly demonstrated that the leaves were the most used by population^{71, 37}. The use of leaves can be explained could be explained by the abundance of phytochemical compounds responsible for the plant's biological properties⁷². Leaves are the place of synthesis of vegetal secondary metabolites⁷².

In order to facilitate and make effective the administration of the active ingredients of plants, several methods of preparation were employed. The finding reveals that incense is the most dominant method of preparation followed by the decoction. The incenses are mostly used for air disinfection to prevent respiratory infectious diseases like common cold and flu⁴⁴. Incense of plants had a good antimicrobial activity against airborne microbes and also this method was easy to prepare, economical and did not lead to health hazards⁷³. While the decoction remains the easiest and most effective way to inactivate or mitigate the harmful and toxic effect of certain plants but can destroy active substances⁷⁴. Many studies have reported that decoction is the most used method for preparing plants^{71, 75}. This result could be explained by the fact that the method of preparation remains linked to the expected objectives of plants. In Morocco, the preparation of plants as against diseases consists mainly of decoction, pasta of herbal, infusion and maceration^{76, 77}.

The results of this study showed that disinfection of rooms and surfaces can be achieved by contact with air or by direct contact with surfaces. These results are corroborate with the French hygiene society which reports that the disinfection of premises, surfaces and medical devices can be done by air (outside or in the presence of humans), by immersion or by direct contact with disinfectant⁷⁸. The antimicrobial effect of Chinese herbal air disinfectant has been demonstrated⁴⁴.

CONCLUSION: This study conducted with herbalists in the cities of Fez and Meknes has identified plants used as bio-disinfectants. These are 13 species belonging to 8 families: *Lavandula*. sp, *Origanum elongatum*, *Artemisia herba-alba*, *Rosmarinus officinalis*, *Eucalyptus*.sp, *Atractylis gummifera* L, *Citrus limon*, *Anastatica hierochuntica*, *Myrtus communis* L, *Rosa damascena*, *Artemisia absinthium* L, *Corrigiola telephiiifolia* et *Tetraclinis articulata*. Leaves and flowers are the most used part of plants while incense is the most formulated mode. As for the instructions for use, the study showed the dominance of aerial disinfection. This result would be of great interest to health professionals to solve the problem of resistant strains and for the pharmaceutical industry to develop effective biological disinfectants that respect the environment. It would be very interesting to continue this work in other Moroccan regions to expand the list of bio disinfectant plants as well as to study of their antimicrobial activities.

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CONFLICTS OF INTEREST: The authors declare that they have no competing interests.

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