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Research Article

AN IN VITRO STUDY ON THE ANTIMICROBIAL EFFECT OF FUMIGATION WITH *JATU-SARJARASADI CHOORNA*

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ABSTRACT

In the present scenario, shooting incidence of airborne diseases is a major public health challenge. Therefore, disinfecting our immediate surroundings is quite important. In Ayurveda, *Dhoopana* is a disinfection method by which drugs of herbal, herbo-mineral or animal origin are used for fumigation. It is a safe, natural and cost-effective method for disinfection. But the effectiveness of *Dhoopana* has to be proved scientifically for the acceptance of the modern society. Therefore, *Jatu-sarjarasadi dhoopa yoga* mentioned in *Ashtanga samgraha sutrasthana* is selected for the study to evaluate its antimicrobial activity. *Jatu, Sarjarasa, Ushira, Sarshapa, Patra, Valaka, Vella, Arushkara, Pura, Arjuna* are the ten ingredients of the *Yoga*, among which *Arushkara* is replaced in this study with its *Abhava dravya Citraka* due to safety reasons. The study was conducted in the procedure room of Prasootitantra Department of Government Ayurveda College Hospital, Tripunithura. The room was fumigated for 30 minutes and kept enclosed for 24 hours. Total microbial colonies were estimated by settle plate method. Petridishes containing culture medium of bacteria and fungi were exposed for 60 minutes in the room before fumigation and 24 hours after fumigation. The sample plates were incubated and total microbial colonies before and after fumigation were counted. The data was statistically analysed using Wilcoxon Signed Rank Test. On statistical analysis, the reduction in total colony count of bacteriae and fungi was found to be significant and showed profound anti-bacterial activity when compared to anti-fungal action. The study concluded that fumigation with *Jatu-sarjarasadi choorna* has anti-microbial effect.

INTRODUCTION

The immediate environment of man comprises of air on which depends all forms of life. There are millions of micro-organisms are present in the air around us. Among which the pathogenic microbes present in the environment are responsible for various health problems. Shooting number of air borne diseases are a major threat to the current world. Most of the respiratory tract infections are acquired by inhaling the air containing the pathogen. Spread of various pathogens in the air adversely affect the population especially those with weaker immunity.

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Covid-19 pandemic is the best example for that. According to the latest statistics of Centers for Disease Control and Prevention (CDC) and World Health Organisation (WHO), the global death rate of major air borne diseases are 6.9 million for Covid-19[1], 1.6 million for tuberculosis[2], 4.8 million for aspergillosis[3] and 1.2 lakh for measles among the children under age of five[4]. Indoor environment conditions also contribute greatly to human wellbeing, as most people spend around 90% of their time indoors, mainly at home or in the workplace and in the course of a day, a person inhales over 15 cubic metres of air. Hence the microbial content of the air one breathes is important, particularly when it contains pathogens. Hence, decontamination of environment is an essential consideration for the control of pathogens.

Air purification is the process of sanitizing the air by neutralizing airborne toxins (gases, bacteria, viral or fungal matter, toxic pathogens etc). In Ayurveda,

features of *Vishayukta vayu* and its purification methods are described in various classics as fumigation of drugs (*Dhoopana*), medicated flags (*Oushada pataka*), sprinkling method (*Prokshana*), medicated musical instruments. *Dhoopana* (fumigation) is one of the important modality among them. In this various medicinal drugs were burnt on fire and the smoke generated from it is used to make disinfection of different areas where chances of infections are more. Since Vedic period, *Dhoopana* is used to disinfect *Bheshajagara, Vranagara, Sutikagara, Shastrakarma ghruha, Kumaragara* etc. *Dhoopana karma* kills microbes and creates an aseptic environment thus prevents infection. Some formulations are also used to treat *Graharoga, Jwara, Arsha, Unmada, Romantika* and *Shotha* etc., to heal *Vrana, Yonivyapada, Karnarogas, Nasarogas, Gudarogas, Gatradaurgandhya* etc. *Dhoopana* has also been followed as a tradition in various religious procedures. *Rakshoghna vidhi* is also indicated in our classics. Plants which have *Kusthahara, Krimighna* and *Vranahara gana* have mostly been used for their antimicrobial properties. Fumigation with such drugs are safe. It helps to create an aseptic environment without any adverse effects unlike various chemicals and aerosols. Prolonged exposure of the most common chemical disinfectant, formaldehyde can cause breathing difficulties, skin irritation, itching, dermatitis and renal problems. Fumigation is also a natural and cost effective method which makes the environment disease-free. But unfortunately, researches conducted in this field are negligible. Even though, there are impressive effects by the conventional fumigation, claims of Ayurveda have to be proved scientifically for the acceptance of public. Hence it is the need of the hour to think about new formulations to control infectious agents in terms of preventive aspect of diseases**.** The present study evaluated the anti-microbial effect of *Jatu-sarjarasadi dhoopa choorna* which is mentioned by *Vagbhata acharya* in *Ashtanga Samgraha* as one of the *Vishaghna dhoopa* for the disinfection of *Vasagruha.*[5]

AIM

To study the anti-microbial effect of fumigation.

OBJECTIVES

To evaluate the microbial load in air before and after fumigation with *Jatu-sarjarasadi choorna* using settle plate method.

METHODOLOGY

A. Experimental design: Anti-microbial study

B. Study setting: Experimental study was conducted at the procedure room of Prasootitantra department of Govt. Ayurveda College Hospital, Tripunithura. The counting of settle plates was conducted at PVT Meritbiolabs (NABL accredited), Palarivattom.

MATERIALS AND METHODS

Preparation of *Jatu-sarjarasadi choorna*

Ten herbs were used in the preparation of *Jatusarjarasadi choorna*, among which *Arushkara* is replaced with *Citraka* for safety reasons. Raw drugs were purchased from the authentic sources, washed and dried. The ingredients were taken in equal quantity and coarse powdered (mesh size 40-60) separately. A homogenous mixture of *Dhoopa choorna* was prepared by mixing all.

Table 1: Ingredients of *Jatu-sarjarasadi choorna*

S.No	Sanskrit Name	Malayalam Name	Scientific Name	Family	Parts used
1.	*Jatu*	*Kolarakku*	*Laccifer lacca*	Coccideae	Lac resin
2.	*Sarjarasa*	*Chenjalyam*	*Shorea robusta*	Dipterocarpaceae	Oleogum, Resin
3.	*Ushira*	*Ramacham*	*Vetiveria zizanioides*	Poaceae	Roots
4.	*Sarshapa*	*Vellakaduku*	*Brassica campestris*	Cruciferae	Seeds
5.	*Patra*	*Pachila*	*Cinnamomum tamala*	Lauraceae	Leaves
6.	*Valakam*	*Iruveli*	*Pavonia odorata*	Malvaceae	Roots
7.	*Vella*	*Vizhalari*	*Embelia ribes*	Myrsinaceae	Fruits
8.	*Chitraka*	*Koduveli*	*Plumbago zeylanica*	Plumbaginaceae	Roots
9.	*Pura*	*Guggulu*	*Commiphora mukul*	Burseraceae	Gum resin
10.	*Arjunam*	*Neermarutu*	*Terminalia arjuna*	Combretaceae	Flowers, Bark

Media and Reagents

Plate count agar (PCA) was used as the medium for bacteria and Dichloran rose-bengal chloramphenicol agar (DRBCA) was used as the medium for fungus.

Preparation of medium

Plate count agar (PCA)[6]

Table 2: Contents of Plate count agar

Ingredients	Gms/L
Enzymatic Digest of Casein/tryptone	5.0
Yeast Extract	2.5
Glucose	1.0
Agar	15.0

- Suspend 23.5 grams of ingredients in 1000ml distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.
- Cool to 45-50°C.
- Mix well and pour into sterile Petri plates.

Dichloran Rose-Bengal Chloramphenicol Agar **(**DRBCA**)[7]**

Table 3: Contents of Dichloran Rose-Bengal Chloramphenicol Agar

Ingredients	Gms/L
Peptone	5g
Dextrose (Glucose)	10g
Potassium dihydrogen phosphate	1g
Magnesium sulphate	0.5g
Rose Bengal	0.025g
Dichloran	0.002g
Chloramphenicol	0.100g
Agar	15g

- Add the components except for agar and chloramphenicol in the required quantities in 800ml of distilled water.
- Adjust the pH and make up the volume to 1L.
- Add agar
- Autoclave
- Add chloramphenicol while the media is cooling down
- Pour in sterile petri plates

Method of Experimental Study –Settle Plate Method

Fumigation with *Jatusarjarasadi choorna* was done at the procedure room of Prasoothitantra department, Government Ayurveda College Hospital, Thripunithura. Each Petridish of diameter 9cm containing culture medium for bacteria and fungus was left open in the procedure room at a distance of one meter above the floor level for 1 hour. After one hour, the lids of petri plates were replaced and kept in incubator at 37°C. Thereafter fumigation was done. The amount of *Dhoopa choorna* was fixed as 3.3gm/m^2 and the *Choorna* was ignited by using charcoal. Entire room was fumigated for 30 minutes and thereafter, the room was kept enclosed for 24 hours. After 24 hours, petridish containing culture medium was exposed in the room for 1 hour and the lids were replaced after 1 hour and kept in incubator at 37°C.The same procedure was repeated once a week for the next 2 consecutive weeks. In each trial, 5 samples were taken and therefore the total number of samples was 15. The incubation time for bacteria was 48 hours and that for fungus was five days. After incubation, the number of microbial colonies in each set of plates were counted using colony counter and the values were recorded.

Assessment of Efficacy

The efficacy was assessed by the reduction in the mean value of total bacterial and fungal count (in cfu) before and after fumigation.

Collection of Data: Data was collected using settle plate method.

Quality Control and Quality Assurance

The study drugs were purchased from authentic sources, were washed and dried. The *Choorna* was prepared as per classical methods and according to Ayurveda Pharmacopia of India and Ayurvedic Formulary of India.

Ethical Consideration

Approval from the Institutional Ethical Committee, Government Ayurveda College, Tripunithura, was obtained, dated 05-08-2021 (02/SV/IEC/2021) and the study was conducted strictly by following the guidelines of committee.

OBSERVATION

In this study, 3 trials of fumigation were conducted in the procedure room of Prasoothitantra department. In each trial, bacterial and fungal count before and after fumigation were noted. To find out the efficacy of the fumigation, the reduction in the total number of colonies before and after fumigation were considered as the outcome measurement. Since the distribution of the total number of colonies was not following normal distribution, the comparison of the total number of bacterial and fungal colonies before and after fumigation was done by Non-parametric methods. As this was a pre-post comparison, Wilcoxon's signed rank test was used for testing the significance of the reduction in the number of bacterial and fungal colonies. A two tailed p value of 0.05 is considered for significance.

Table 4: Total Number of Bacterial Colonies Before and After Fumigation

	Sample No.	Before Fumigation	After Fumigation
1st Week	1	30	5
	2	11	3
	3	26	4
	4	16	3
	5	22	4
	Mean	21	3.8
2nd Week	6	13	4
	7	18	5
	8	12	3
	9	20	3
	10	10	3
	Mean	14.6	3.6
3rd Week	11	34	18
	12	22	5
	13	20	6
	14	25	4
	15	15	3
	Mean	23.2	7.2
Total Mean Value		19.6	4.8

In first week, the mean value of total bacterial count before fumigation was 21 which is reduced to 3.8 after fumigation. In the second week, the mean value before fumigation was 14.6 which is reduced to 3.6 after fumigation. In the third week, the mean value reduced from 23.2 to 7.2 after fumigation. The total mean value of 15 samples before fumigation was 19.6 which reduced to 4.8 after fumigation.

Figure 3: Comparison of Total Number of Bacterial Colonies before and After Fumigation

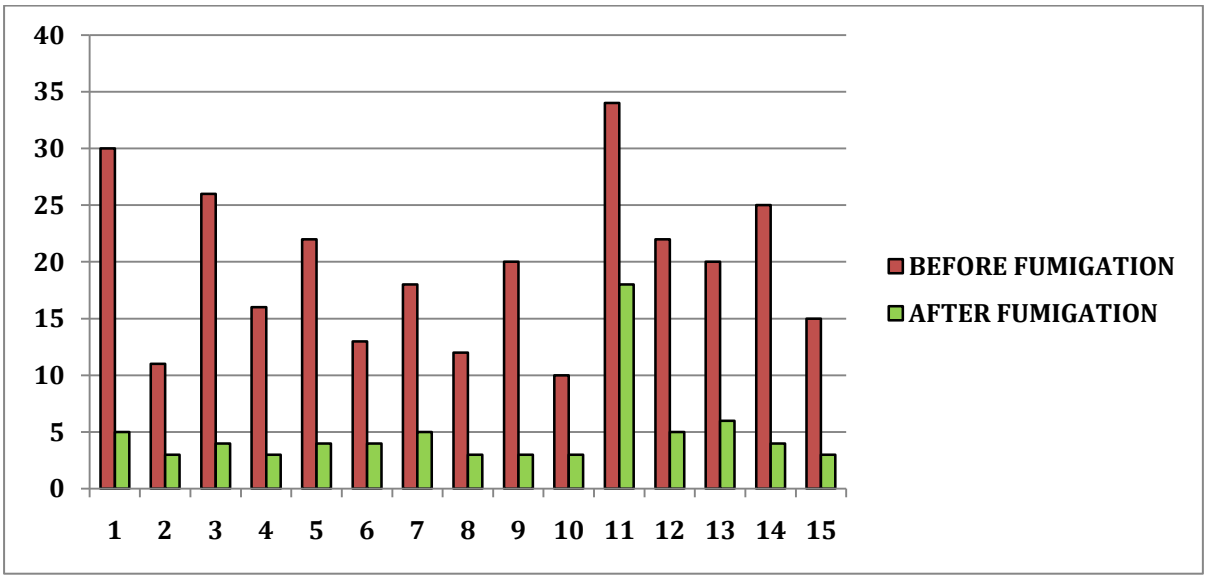

Table 5: Comparison of Total Number of Bacterial Colonies Before and After Fumigation

Total number of bacterial colonies	Before fumigation	After fumigation	Wilcoxon's Signed Rank Test		Significance
			Z value	P value	
Mean	19.6	4.866	-3.407	0.000	Highly Significant
Median	20.0	4.0			
S.D	7.079	3.758			

Before fumigation, the mean count of total number of bacterial colonies was 19.6 which are reduced to 4.8 after fumigation. Similarly, the median is reduced from 20.0 to 4.0 by the process of fumigation. The Wilcoxon's Signed Rank Test (Z value=3.407) and p value (0.000) shows that the reduction in the total number of bacterial colonies is highly significant.

Table 6: Total Number of Fungal Colonies Before and After Fumigation

	Sample No.	Before Fumigation	After Fumigation
1st Week	1	8	6
	2	4	5
	3	7	4
	4	9	5
	5	8	10
	Mean	7.2	6
2nd Week	6	7	3
	7	8	4
	8	11	5
	9	9	11
	10	10	6
	Mean	9	5.8
3rd Week	11	18	6
	12	21	11
	13	19	8
	14	13	6
	15	11	13
	Mean	16.4	8.8
Total Mean Value		10.8	6.8

In the first week, the mean value of total fungal count before fumigation was 7.2 which was reduced to 6 after fumigation. In the second week, the mean value before fumigation was 9 which reduced to 5.8 after fumigation. In the third week, the mean value reduced from 16.4 to 8.8 after fumigation. The total mean value of 15 samples before fumigation was 10.8 which reduced to 6.8 after fumigation.

Figure 6: Comparison of Total Number of Fungal Colonies Before and After Fumigation

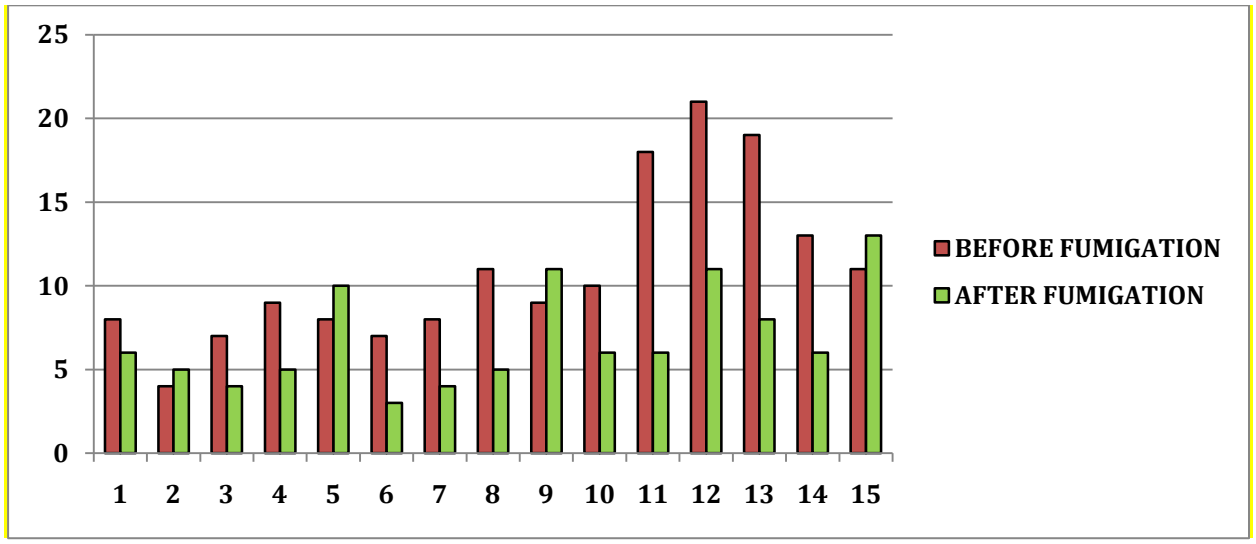

Table 7: Comparison of Total Number of Fungal Colonies Before and After Fumigation

Total number of bacterial colonies	Before fumigation	After fumigation	Wilcoxon's Signed Rank Test		Significance
			Z value	P value	
Mean	10.866	6.866	-2.754	0.005	Significant
Median	9	6			
S.D	4.882	3.020			

Before fumigation, the mean count of total number of fungal colonies was 10.8 which reduced to 6.8 after fumigation. Similarly, the median reduced from 9.0 to 6.0 by the process of fumigation. The Wilcoxon's Signed Rank Test (-2.754) and p value (0.005) shows that the reduction in the total number of fungal colonies is significant.

Petri plates containing Bacterial Colonies Before and After Fumigation

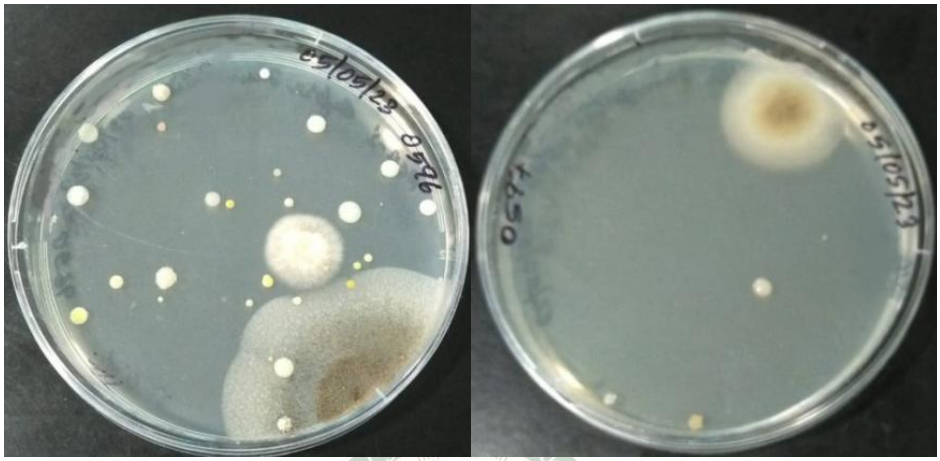

Figure 1: Before fumigation [Sample 1] Figure 2: After fumigation

Petri plates containing Fungal Colonies Before and After Fumigation

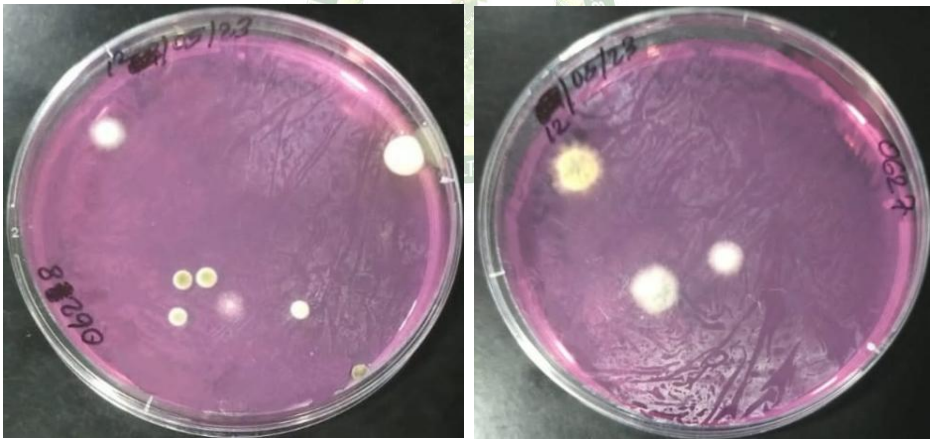

Figure 4: Before fumigation [Sample 6] Figure 5: After fumigation

DISCUSSION

Jatusarjarasadi dhoopana yoga is mentioned by *Vagbhata* in *Annaraksha vidhi adhyaya* in *Ashtanga samgraha sootrasthana* to fumigate the places of dwelling. *Jatu, Sarjarasa, Ushira, Sarshapa, Patra, Valaka, Vella, Arushkara, Pura, Arjuna* are the ten ingredients of the *yoga*. All drugs are taken in equal quantity for the preparation of *choorna* with mesh size 40-60. These drugs are having antimicrobial properties mentioned as *Bhutaghna, Krimighna, Vishaghna* in Ayurvedic *Samhitas*. As it contains 10 potent drugs, this formulation has a wider range of utility than other small *Dhoopana yogas*. It is a formulation indicated to remove poisons of both plant and animal origin and act as a repellent of insects, rodents and reptiles. All ingredients are easily available in the market except *Arjuna pushpa*. So the study was carried out by using the bark of *Arjuna* and still showed significant result. A previous study of antimicrobial activity on the methanolic extract of *Arjuna* bark showed significant zones of inhibition against twenty-two tested bacteria and eight uropathogens[8].

One of the ingredients *Bhallataka* is reported under *Upavisha Dravya* in classical Ayurvedic texts. Its fruit contains anacardiac acid, cardol, catechol, anacardol and a fixed oil, somecarpol and Bhilawanol. The Phenolic compounds are thought to be toxic to micro- organisms, inhibiting the enzymes which are

essential for the growth of microorganism. As per expert opinion, even though it has bacteriostatic property, it needs more safety precautions to handle with the fumes of raw fruit. The fruit containing oil is blistering and yield 32 percent. It is soluble in ether and it turns black by air contact. A previous study shows that 'urushiol' is the active allergen found in the smoke of *Semecarpus anacardium* Linn seeds which is responsible for the allergic contact dermatitis in patients exposed to the smoke of *Semecarpus anacardium* Linn seeds[9]. So, the drug is replaced with its *Abhava dravya citraka* as mentioned in *Bhavaprakasha* which is similar in properties[10]. And still the study showed significant antimicrobial effect. The formulation is also economically feasible as it does not contain any expensive drugs. Hence this *Dhoopana yoga* is selected for the study.

While analyzing literature review of the anti-microbial activity of ten ingredients in *Jatusarjarasadi dhooma choorna*, eight of them shows significant anti-bacterial property especially against gram positive bacteria and only 3 among the ingredients (*Vella, Citraka* and *Arjuna*) shows significant anti-fungal activity and 2 of them (*Valaka* and *Sarja rasa*) have moderate anti-fungal activity.

***Jatu*:** Solvent wise variations in antimicrobial activity of lac were observed against different test microbes. Acetone extract of lac showed antimicrobial activity against five microbes i.e., Escherichia coli, Staphylococcus aureus, Bacillus subtilis, Xanthomonas citri and Erwinia carotovora, the highest being against Escherichia coli. It showed moderate activity against Erwinia carotovora and Xanthomonas citri and lesser antimicrobial activity against Bacillus subtilis and Staphylococcus aureus[11]. This proves the antimicrobial activity of *Jatu*.

***Sarja rasa*:** *Shorea robusta* resin has a stronger and broader spectrum of antimicrobial activity against a number of pathogenic microorganisms. Aqueous extracts of *Shorea robusta* exhibits significant activity against Bacillus coagulans, Escherichia coli, Bacillus cereus and moderate inhibition on Salmonella typhi and Bacillus subtilis and less activity against Proteus vulgaris and Pseudomonas fluorescence.

However, ethanolic extracts also exhibited significant activity against Staphylococcus aureus, S. epidermidis and Escherichia coli, moderate inhibition on Candida albicans and Bacillus coagulans. The petroleum ether extract showed activity against Escherichia coli, Aspergillus flavus and Candida albicans and whereas benzene extracts worked against Bacillus licheniformis, Bacillus cereus and Aspergillus flavus[12]. This proves the antimicrobial activity of *Sarja rasa*.

***Usira*:** Anti microbial activity of *Vetiveria zizanioides* (vetiver) against Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli and Corynebacterium ovis were evaluated. Against S.aureus, vetiver oil was superior to the other two oils in the pure state and diluted with dimethyl sulphoxide 1:10, 1:100, 1:1000 and 1:10000; inhibition by the pure oil was 60-70% that by penicillin or streptomycin[13].

***Sarsapa*:** A study to investigate how mustard essential oil (EO) affected the cell membrane of Escherichia coli O157:H7 and Salmonella typhi. Intracellular pH and ATP concentration and the release of cell constituents were measured when mustard EO was in contact with E. coli and S. typhi at its minimal inhibitory concentration (MIC) and maximal tolerated concentration (MTC). The treatment with mustard EO affected the membrane integrity of bacteria and induced a decrease of the intracellular ATP concentration. Electron microscopy observations showed that the cell membranes of both bacteria were apparently damaged by mustard EO[14]. This proves the antimicrobial activity of mustard essential oil.

***Patra*:** In-vitro anti-microbial potential of *Cinnamomum tamala* leaves extracts showed variable degree of inhibition zones against the selected six gram negative, three gram-positive bacterial strains and a fungus. All the extracts showed their best inhibitory activity against B. atrophaeus, amongst which the aqueous extract recorded the highest zone of inhibition measuring 38mm[15]. This proves the anti-microbial activity of *Patra*.

***Valaka*:** The essential oil of *Plectranthus vettiveroides*, the South Indian variety showed anti bacterial activity against Staphylococcus aureus, Escherichia coli and Pseudomonas aerogenosa comparable to amoxicillin and antifungal activity against Candida albicans and Penicillium notatum comparable to mycostatin[16]. This proves the antimicrobial activity of *Valaka*.

***Vella*:** Antifungal activity of *Embelia ribes* was evaluated on eight different fungal species by employing various concentrations of seed extract (0.5-2.0mg). Among all the fungi, high inhibition zones were observed in *Colletotricum crassipes* (18mm). This was followed by Cladosporium (17.5mm), *Armillaria mellea* (17mm), *Colletotricum capsici* (17mm), *Aspergillus niger* (16.5mm), Rhizopus oryzae (16.5mm), respectively. *Aspergillus terreus* and Candida albicans showed less inhibition zones (15.5 and 16.0mm) compared to other organisms[17]. It clearly demonstrated the antifungal properties of *Embelia ribes*.

With regard to antibacterial activity, embelin showed bactericidal activity against Gram +ve organisms, and bacteriostatic against Gram -ve organisms[18].

***Citraka*:** The methanolic extracts of the stem and the leaves of *Plumbago zeylanica* were tested against six bacterial species and nine fungal species, and both extracts showed antimicrobial activity in a dose-dependent manner. The leaf extract of Plumbago

zeylanica showed maximum antimicrobial activity against both Staphylococcus aureus sub sp aureus and Fusarium oxysporum. The stem extract was found to be more antimicrobial against the Pseudomonas aeruginosa and the Penicillium expansum species[19]. This proves the anti microbial activity of *Citraka*.

***Pura*:** Extract of *Guggulu* gum possesses significant antibacterial activity against gram-positive bacteria and moderate activity against gram-negative once. S. aureus and S. aglactiae were found to be most susceptible organisms whereas E. coli was shown resistance and no inhibition reported for the P.aerugunosa[20]. This validates the antimicrobial activity of *Guggulu*.

***Arjuna*:** The water and methanol extracts of T. *Arjuna* bark produced significant zones of inhibition against twenty-two tested bacteria including eight uropathogens. MIC values against the bacteria were found in the range of 0.16 to 2.56mg/mL. The polar extracts of T. *Arjuna* also demonstrated strong anti-fungal effect against eight species of Candida, with MIC between 0.16 and 0.64mg/mL[21]. This validates the antifungal activity of *Arjuna*.

Most of the ingredients have significant anti-bacterial action than anti-fungal property. So, this may be the reason behind the disparity in the result of total bacterial and fungal count. However, the formulation consists of drugs that show a synergetic effect of anti-bacterial and anti-fungal activity. So collectively this is useful to generate smoke which exerts its anti-microbial action.

Probable Mode of Action of Fumigation

Generally *Dhoopana yoga* contains aromatic plants as well as resins to bind it together. In *Jatusarjarasadi Dhooma choorna, Tamalapatra, Guggulu, Sarja, Ushira* are aromatic while *Laksha, Guggulu, Sarja* contains binding resins. All these drugs are having resins, oleoresins, phenolic compounds, flavonoids, volatile oils and essential oils. After burning the *Dhooma choorna*, volatile principles are released in air. Some studies have indicated that adding volatile oils to meals delay microbiological infection, will prevent the loss of organoleptic qualities and hence delay the start of spoiling[22]. And also it is proven that the volatile oils present in the fumes have the capacity to damage the cell integrity of bacteria[23]. Most of the ingredients of *Jatusarjarasadi Dhooma choorna* have *Katu, Tikta* and *Kashaya rasa, Teekshna guna* with *Ushna veerya*. They have predominance of *Vayu mahabhoota* along *with Agni, Aakash* and *Prithvi mahabhoota*. *Vayu* and *Akasha* are responsible for quicker combustion and rapid spread of smoke[24]. Due to predominance of *Vayu, Agni* and *Akasha*, the smoke has tendency to move upward and spreads quickly everywhere. Due to hot property of ingredients the temperature of atmosphere increases which is opposite to damp and humid conditions that is favourable for microbes to grow in general.

CONCLUSION

In the present world, upcoming airborne infections are a major global health concern. The population is negatively impacted by the spread of numerous infections in the air, especially individuals with weaker immunity. The Covid-19 epidemic is the prime example for that. Decontamination of environment is an essential consideration for the control of pathogens and prevention of communicable diseases. Ayurveda promote disease prevention through a variety of techniques. *Dhoopana* (fumigation) is one of the important modality among them which is a safe, natural and cost effective method of disinfection. While traditional fumigation can produce impressive results, Ayurvedic claims need to be scientifically proven in order to be accepted by the general public. Hence, the present study was an attempt to assess the anti-microbial effect of *Jatusarjarasadi dhoopa choorna* which is mentioned by *Vagbhata acharya* in *Ashtanga Samgraha* as one of the *Vishaghna dhoopa* for the disinfection of environment. The study evaluated the microbial load in air before and after fumigation with *Jatusarjarasadi choorna* using settle plate method in the procedure room of Prasootitantra department of Government Ayurveda College Hospital, Thripunithura. The fumigation with Jat*usarjarasadi choorna* showed significant anti-microbial effect. The anti-bacterial activity of fumigation with *Jatusarjarasadi choorna* was highly significant compared to its anti-fungal activity.

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