

## **A Comparative Study of *Arishta Dhoopana Yoga* and Formalin for their Antimicrobial Effect as a Fumigating agent on *Escherchia Coli***

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### **Abstract**

#### **Background:**

Fumigation is a procedure to prevent and control the Nosocomial infections among the patients and staff. Nosocomial infections or hospital acquired infections are defined as infections acquired during or as a result of hospitalization. It may not be possible to eradicate all hospital related infections. However, an effective infection control program provides optimum protection for both the patients and hospital staff. By maintaining sterile environment in surgical theatre, hospital wards; we can control major part of exogenous infections. Formaldehyde fumigation has been an accepted method of sterilization for areas where microbiological cleanliness is required. Fumigation with formaldehyde vapour is the recognized and most commonly used method because of its cost effective procedure. Chemical agents of fumigation like Formaldehyde are having many hazardous effects on human body. The purpose of this study is to help health care organisations achieve the best possible, cost effective Herbal fumigation (*Arishta Dhoopana Yoga*) infection control measures of specific microorganism (*E. coli*)

#### **Method:**

Fumigation effect of Herbal fumigation (*Arishta Dhoopana Yoga*) and Formalin were compared on *E. coli* (prepared in the form of agar slant) sprayed glass experimental chambers of 1M<sup>3</sup>. The colony forming units (CFUs) of *E. coli* by taking swabs for culture before fumigation (0 Minute) and after fumigation (at 30 minutes, 3 Hours) were noted. The observations of each group and intra group are compared.

#### **Results:**

Significant results are noted in CFUs after fumigation in each group. *Arishta Dhoopana Yogafumigation* showed significant antimicrobial activity.

#### **Interpretation:**

*Arishta Dhoopana Yoga* fumigation showed significant antimicrobial activity as Formalin fumigation. Herbal fumigation (*Arishta Dhoopana Yoga*) is best possible and cost effective infection control measures of specific microorganism (*E. coli*).Herbal fumigation (*Arishta*

*Dhoopan Yoga*) has no hazardous effects on human body. So this Herbal fumigation (*Arishta Dhoopan Yoga*) can be considered for routine hospital fumigations.

### **Keywords**

*Fumigation, Herbal Fumigation, Dhoopan, ArishtaDhoopan, Nosocomial infections, Ayurveda & Fumigation*



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## INTRODUCTION

*Ayurveda* believes in striking a balance between an individual (body, mind, soul, spirit) and environment. Measures like water purification, *Homa-Yagya*, *Dhoopan* has been mentioned in many *Ayurvedic* texts. *Dhoopana* is mentioned by which drugs of herbal, herbo-mineral, mineral, or animal origins are used for fumigation (*Dhoopan*)<sup>1</sup>. Mankind is exposing to number of chemicals every day, from many studies it is observed that the use of chemical above certain limits is hazardous to human lives. Formalin and other fumigating agents may cause - skin rash, eye irritation and having unpleasant odour and cancerous effect<sup>2</sup>. Studies have showed that; Microbial contamination of hospital environment, especially in operation theatre and other specialized units had continued to increase prevalence of nosocomial infections<sup>3</sup>. Air disinfectants are chemical substance capable of disinfecting micro organisms suspended in air<sup>4</sup>. *Acharya Kashyap* has described 40 types of *dhoopas* in *Kalpasthan Dhoom kalpadhyay* for various diseases & various diseased conditions. *Kashyapa* has described the *Arishta Dhoopana Yoga* for the fumigation of *Krumi*, *Bhoota*, *Pishaccha* (micro organisms)<sup>5</sup>. *Neem* is extendedly used in *Ayurveda*, *Homeopathy* and *Unani*

medicine and has become cynosure of modern medicine<sup>6, 7</sup>. The Sanskrit name of *Neem* is '*Arishta*' meaning 'reliever of sicknesses' and hence considered as '*Sarvroganivarani*'<sup>6</sup>. *Escherichia coli* a gram negative, non sporing, non capsulated, rod shaped bacteria commonly present in lower intestine of human and mammals, and most commonly occurs in hospital environment<sup>8</sup>. *Neem* has broad spectrum antimicrobial & antiseptic activity; is widely used for the management of diseases in *Ayurveda*<sup>6,7</sup>; so that considered for its antimicrobial activity on *E. coli* with *Panchang Dhoopan* (fumigation) for this study. Fumigation (*Dhoopan*) should be done repeatedly as one time fumigation doesn't give protection; repeated fumigation should be done to avoid repeated infections of the patient<sup>5</sup>.

## AIM

To Study Comparative Effect of '*Arishta Dhoopana Yoga*' and Formalin for fumigation on *E.coli*

## OBJECTIVES

- To evaluate efficacy of *Arishta Dhoopana Yoga* as fumigating agent on *E. coli*.

- To compare the antimicrobial effect of Formalin and *Arishta Dhoopana Yoga* on *E. coli*.
- To develop standard protocol for fumigation with *Arishta Dhoopana Yoga* as an alternative.

## MATERIALS

- **NeemPanchang Powder**<sup>7,9</sup>:

**Latin name:** *Azadirachta indica* A. Juss

**Family:** Meliaceae

**Chemical composition:** Triterpenoids, limonoids, azadirachtin, azadiradione, nimbin, nimbolide, nimbidin, nimbinin, sitosterol, margosinolide, tocopherol, salannol, Sterols, margosine, volatile oils, astringent elements, gum, sugar, white secretion and traces of sulphur.

**Part used:** *panchangas* **Rasa:** *Tikta, Kashaya* **Guna:** *Laghu* **Veerya:** *Katu* **Vipaka:** *Sheeta* **Doshakarma:** *Kapha, Pitta Shamaka* **Karma:** *Krumighna, Kusthagna, Kandughna, Vishaghna, Vranaghna, etc.*

- Two experimental chambers of 1meter cube made up of glasswere constructed,
- *Escherichia Coli* (non-pathogenic organisms) in agar slant form.

- It is an experimental study conducted in one meter cube experimental chamber made up of glass.

- Non-pathogenic *E.coli* organism is prepared in the form of agar slant for fumigation with standard method.

- Firstly cleaning of Trial and Control experimental chambers was done.

- Each ingredient of *Arishta Dhoopana Yogawas* taken in equal quantity<sup>11</sup>.

- Standardization and authentication of *Arishta Dhoopana Yoga* was done from authentic source; which was found standard.Preparation of *Arishta Dhoopana Yoga* was done with standard operating procedure guidelines.

- Then the spraying of *E.coli* organism in both experimental chambers was done and pre fumigation swabs were taken from each experimental chambers walls and air.

- Fumigation with '*Arishta Dhoopana Yoga*' in Trial Experimental chamber and Formalin in Control Experimental chamber was performed.

- Post fumigation swabs were taken after 30 minutes and 3 hours from both experimental chambers. Swabs were taken from roof, floor, wall and air.

## METHODS

- Swabs sent to microbiology lab for testing; the observations were recorded, analysed and compared accordingly.

## OBSERVATIONS

The culture swabs of pre-fumigation (0 min) and post fumigation (after 30min and 3 hours) obtained, sent for culture and the observations (colony count) were noted. The observations are as below:

### TRIAL EXPERIMENTAL CHAMBER OBSERVATION:

**Table 1** Observation of fumigation with *Arishtadhoopan* on *E.coli*:

	Pre-fumigation (0min)					Post -fumigation (30min)					Post-fumigation (3hrs)				
	1cfu	2cfu	3cfu	4cfu	5cfu	1cfu	2cfu	3cfu	4cfu	5cfu	1cfu	2cfu	3cfu	4cfu	5cfu
<b>Date</b>	23sept 2016	24sept 2016	26sept 2016	27sept 2016	28sept 2016	23sept 2016	24sept 2016	26sept 2016	27sept 2016	28sept 2016	23sept 2016	24sept 2016	26sept 2016	27sept 2016	28sept 2016
<b>Roof</b>	74	68	54	62	80	65	63	48	54	72	52	48	42	50	67
<b>floor</b>	88	76	72	80	84	81	70	68	75	79	68	58	52	68	61
<b>Wall</b>	90	110	105	86	92	85	91	90	78	85	60	72	81	65	75
<b>Air</b>	128	120	132	122	115	105	97	110	82	92	90	80	92	72	82

### CONTROL EXPERIMENTAL CHAMBER OBSERVATION:

**Table 2** Observation of fumigation with Formalin on *E. coli*

	Pre-fumigation (0min)					Post fumigation (30min)					Post-fumigation (3hrs)				
	1cfu	2cfu	3cfu	4cfu	5cfu	1cfu	2cfu	3cfu	4cfu	5cfu	1cfu	2cfu	3cfu	4cfu	5cfu
<b>Date</b>	23sept 2016	24sept 2016	26sept 2016	27sept 2016	28sept 2016	23sept 2016	24sept 2016	26sept 2016	27sept 2016	28sept 2016	23sept 2016	24sept 2016	26sept 2016	27sept 2016	28sept 2016
<b>Roof</b>	154	123	127	165	146	104	121	108	102	98	91	87	79	85	76
<b>Floor</b>	134	142	154	136	98	96	129	132	101	84	64	103	81	85	62
<b>wall</b>	120	153	148	125	138	94	120	108	110	121	65	93	80	102	100
<b>Air</b>	139	128	143	106	117	90	115	102	75	94	69	65	56	85	78

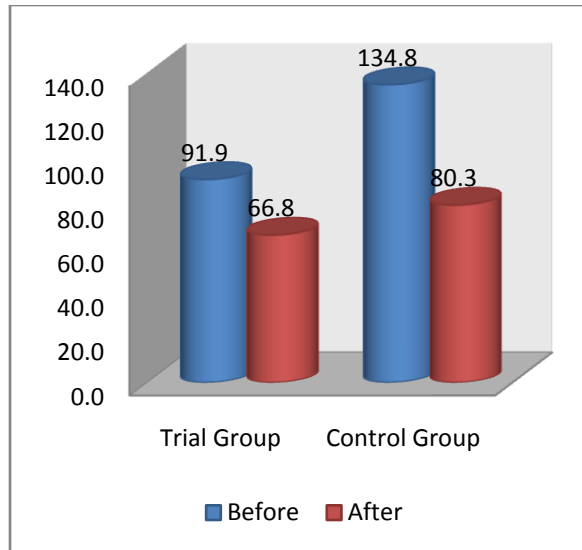
## EFFECT OF TRIAL AND CONTROL GROUP AFTER FUMIGATION

**Table 3** Effect of fumigation on both the groups:

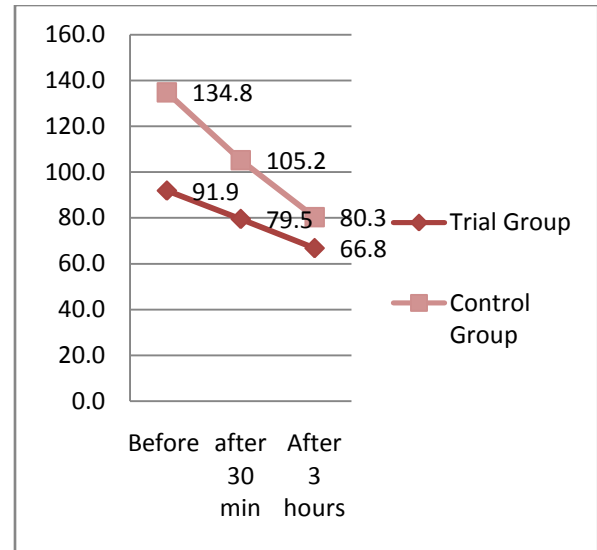
Escherichia coli	Mean		t-Value	P-Value	Result
	Before	After			
Trial Group	91.9	66.8	10.069	0.000	Significant
Control Group	134.8	80.3	12.947	0.000	Significant

Paired t-test used to test the effect of Trial Group and Control Group. From above table it is observed that P-Value of Trial

Group and Control Group are less than 0.05 hence it is concluded that effect observed in both groups are significant.



**Graph No. 1:** showing effect of fumigation in both groups



**Graph No 2** showing effect of fumigation in both groups

**Table 4** Intra groups comparison

Group	N	Mean Diff	SD	SE	t-Value	P-Value
Trial Group	20	25.2	11.17	2.50	-5.996	0.000
Control Group	20	54.5	18.82	4.21		

### COMPARISON BETWEEN PERCENTAGE CHANGE IN BEFORE AND AFTER FUMIGATION

**Table 5** Percentage change before and after fumigation

E. coli	Mean			
	Before	after 30 min	After 3 hours	% Change
Trial Group	91.9	79.5	66.8	27.4
Control Group	134.8	105.2	80.3	40.4

From above table it is observed that P-Value is less than 0.05 which concludes that there is significant difference between effect of trial group and control group. Further mean difference observed in control group is more hence control group is more effective than trial group.

## RESULTS

Comparing the effect of *Arishta Dhoopana Yoga* and Formalin for their antimicrobial effect on *E. coli*, the percentile reduction

before and after fumigation in Trial and Control group are 27.4 and 40.4, respectively. The effect of *Arishta Dhoopana Yoga* on *E. coli* colonies reduction is effective after 30 minute but more effect observed after 3 hours.

## DISCUSSION

*Nimba (Azadirachta indica)* is a medicinal plant having wide spectrum of biological activity and proven as an antimicrobial. *Dhoopana* should be given with the help of

Powder(*churna*) of *Arishtapatra*, *mula*, *pushpa*, *phala*, *twaka* & *Ghrita* (Cow Ghee). The efficacy of *Arishta Dhoopana* drug may prove beneficial as fumigating agents to overcome the ill/adverse-effects of modern techniques of fumigation<sup>9</sup>.

*Neem* has therapeutics implication in the disease prevention and treatment; it is considered that *Azadirachta indica* shows therapeutic role due to the rich source of antioxidant and other active components. *Neem* plants parts shows antimicrobial role through inhibitory effect on microbial growth/ potentiality of cell wall breakdown. Azadirachtin a complex tetranortriterpenoid limonoid present in seeds is the key constituent responsible for both anti feedant and toxic effects in insect. Results suggest that ethanol extract of *Neem* leaves shows in vitro antibacterial activity<sup>8</sup>.

Antibacterial activity revealed that *Neem* leaves extract (50 mg, 75 mg) inhibited the growth of *S. aureus*, *Corynebacterium bovis* and *E. coli*. These results were also supported by Kheret al. (1984) they reported that 10% chloroform extract of *Neem* imported inhibitory effect against *Staphylococcus aureus* and *E. Coli*<sup>3</sup>. *Arishta Dhoopana Yoga* is easily available, easy to prepare, cheap and eco-friendly.

The *E. coli* is selected for the study because it is the common micro organisms that occur in hospital environment, easy availability of cultures, rapid ability to grow colonies, required same nutrient broth for culturing and minimal incubation period and temperature<sup>8,12</sup>.

The outcome of this study is the *Arishta Dhoopana Yoga* proved its antimicrobial activity; so it can be used as an alternative for fumigation in hospital wards/Operation Theatre; as other chemical agents of fumigation are having many hazardous effects on human body.

The *Arishta Dhoop* (fumigation with *Neem* parts and Cow Ghee) having no side effects, which is easily available and very cost effective can be considered for fumigation which was widely used by ancient Ayurveda Scholars for the management and control of various diseases. This concept of herbo-mineral compound fumigation has been described by ancient scholars to control the disease process in *Balagraha* (various systemic infections) management<sup>14</sup>. Apart from the significant physical & medical applications like cleansing of the environment, curing bodily ailments & augmenting vitality and physical potentials, *Dhoopan* is also found to be of immense use in treatment of psychosomatic disorders

& *Balagrahas*, psychological and psychiatric problems<sup>14</sup>.

One of the main ingredients of the *dhoopan karma* is Cow's ghee (Clarified butter) which has enormous beneficial properties. The ghee when burnt with medicinal plants; produces natural fumes which heal the respiratory system, clear blood clots, bacterium affecting the nasal mucosa, lungs & veins.<sup>15</sup>

The quantity of drug required for fumigation of one meter cube area is observed as 40grams. The minimum time required for the sterilization is observed about 30 minutes; but more positive outcomes are observed after 3hours of fumigation. The fumigation (*Dhoopan*) should be done repeatedly; as the one time fumigation doesn't give the protection. The repeated fumigation should be done to avoid the repeated infections of the patients. Hence repeated and prolonged fumigation with *ArishtaDhoop* should be considered to control the Noscomial infections.

## CONCLUSION

*Arishta Dhoopana Yoga* is moderately efficient fumigant against the micro-organism *Escherichia coli*. The action of

*Arishta Dhoopana Yoga* on *E. coli* proved to be anti-microbial. The quantity of *Arishta Dhoopana Yoga* required for fumigation of experimental chamber of one meter cube is 40gm. The time required for the sterilization is 30min, but more effectively sterilization was done after 3hours. The percentile reduction of pre fumigation and post fumigation of Trial experimental chamber (*E. coli*) was significant.



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