INNSPUB

International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print), 2222-5234 (Online)
http://www.innspub.net
Vol. 8, No. 5, p. 96-104, 2016

RESEARCH PAPER OPEN ACCESS

The potential of some spice extracts for controlling *Aspergillus* species

Wajiha Seerat[1*], Abida Akram[1], Rahmatullah Qureshi[1], Javaid Asad[2], Shaista Akhund[1], Brian Gagosh Nayyar[1]

[1]*Department of Botany, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi, Pakistan*

[2]*Department of Biochemistry, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi, Pakistan*

Key words: Antifungal assay, Spice, *Aspergillus*spp., Inhibitory effects, Methanolic extracts.

http://dx.doi.org/10.12692/ijb/8.5.96-104 Article published on May 18, 2016

Abstract

The methanolic extracts of 30 spices were screened for the potential antifungal activity against *Aspergillus niger* and *Aspergillus oryzae*at 25, 50 and 75% concentrations. The extracts of spices were subjected to antifungal assays with the help of two techniques i.e. Poisoned Food Technique and Agar Well Diffusion Method. The maximum percentage inhibition of both the tested fungal strainswas observed at 75% concentrationwhile at 25% concentration, there was minimum inhibition. Overall *Myristica fragrans, Piper nigrum, Cuminum cyminum,* and *Trachyspermum ammi* extracts showed the highest antifungal activity. This study revealed that spices possess good antifungal activity that can be used in many herbal formulations to cure infections.

* **Corresponding Author:** Wajiha Seerat ✉ wajihaseerat@gmail.com

Introduction

Medicinal plants constitute a rich source of antimicrobial agents (Mahesh andSatish, 2008). In traditional medicine, many ofthe plant materials used are readily available in rural areas at relatively cheaper than modern medicine (Mann *et al.,* 2008). Plants generally produce many secondary metabolites which constitute an important source of pesticides, microbicides and many pharmaceutical drugs. Plant products still remain the mainsource of pharmaceutical agents used in traditional medicine (Ibrahim, 1997;Ogundipe *et al.,* 1998).

Fungal contamination of deposited commodities is a very severe problem in various regions of the world. Contamination by storage fungi and the mycotoxins produced by them is of greatsignificance in food industry and herbal medication.Fungi, especially *Aspergillus* species and *Penicillium* species, are the major causes of food spoilage, especially intermediate-moisture food products, preserved grain, bakery products, cheese and fruit.

Wheat bread contamination was mainly *Penicillium* species (90-100%) and also *Aspergillus* species (Legan and Voysey, 1991).

Aspergillus species producing mycotoxins related materials diminish the quality of foodstuffs and the medical value of herbal medications. In previous studies, *Aspergillus niger* commonly recognized as black *Aspergilli*, during storage to be associated with herbal drugs, was noted as a most directing fungal species (Bugno *et al.,* 2006; Gautam and Bhadauria, 2008; Gautam and Bhadauria, 2009).

Plants and their products have been used by humans in various ways and the best use is as food and spices. Many herbs and spices have been used for centuries as preservatives for foods and curative purposes; some of them possess antimicrobial activity in combination and is considered as substitutions to conventional antimicrobial agents especially in this period of antimicrobial drug resistance (Nwaopara, 2009).

Spices have been defined as plant materials from indigenous or exotic origin, aromatic or with strong taste, used to enhance the flavour of foods (Germano and Germano, 1998). Spices include leaves (mint, laurel, bay, coriander, oregano, rosemary), bulbs (onion, garlic), fruits (red chilli, cumin, black pepper), flowers (clove), rhizomes (ginger), stems (cinnamon, coriander), and other plant parts (Shelef, 1983). Although, spices have been well known for their antioxidant, preservative and medicinal properties, they have been currently used with primary purpose of enhancing the taste of foods rather than extending shelf-life (Aktug and Karapinar, 1986; Ristori, 2002).

When spices show initially high microbial control and as time progresses, the microbial growth become gradually slower or it is eventually totally inhibited, this could be observed that spices prevent the infectious deterioration of food (Kizil and Sogut, 2003). There has been maximum concern of the users about foodstuffs free from chemical preservatives or with minor level of chemical additives because these could be noxious for human health (Bedin *et al.,* 1999). The main objective of this study was to investigate the inhibitory effects of methanolic extracts from 30 spices against *A.niger* and*A. oryzae*.

Materials and methods

Collection of plant material

The plant materials of different plant species for this study were collected from local market which was identified by the senior botanist at Department of Botany, PMAS Arid Agriculture University Rawalpindi, Pakistan.

Preparation of extracts

The selected parts of different spices (Table 1) were cut into small pieces and dried in shade at room temperature, and then powdered with the help of grinder. Maceration procedure was adapted for extract preparation. Fifty grams of powdered plant materials were extracted with 150ml of methanol for 3 to 5 days. After 5 - 7 days, the extract was filtered using muslin cloth. The filtrate obtained was again filtered using filter paper. Then rotary evaporator was

used for purification (Penkhae*et al.,* 2005).

Test microorganisms

Two fungal species *viz.Aspergillus niger* and *Aspergillus oryzae* were collected from the Department of Botany, PMAS Arid Agriculture University Rawalpindi. The stock cultures were maintained in Potato Dextrose Agar (PDA) media and 3 – 4 days old cultures of the said fungal pathogens was used for screening.

Screening for antifungal activity

Poisoned food technique

100µl of different concentrations i.e. 25%, 50% and 75% of the methanolic extracts were added in Potato Dextrose Agar (PDA) media. Three replicates of each concentration were maintained. 3 – 4 days old culture of the fungi was used for inoculation. Single spore inoculation was done at the center of each petri plate. Then the plates were incubated at 28°C for 3 to 7 days and diameter of colony formed in each plate was measured and recorded after seven days. PDA plate with methanol was used as negative control. PDA plate with 1000ppm solution of antifungal drug (Terbinafine) was used as positive control. All the results were analyzed statistically (Rajani *et al.,* 2012).

Percent mycelial growth inhibition = ((a-b)/ a) × 100

Where

a = diameter of fungal colony (mean) in control

b = diameter of fungal colony (mean) with plant extract

Agar well diffusion method

PDA plates were swabbed with spore suspension of fungal strains with the help of spreader. 6mm wells were prepared in PDA agar plates by using a sterile cork borer. 100µl of different concentrations i.e. 25%, 50% and 75% of the methanolic extracts were poured into each well. A sample well with methanol was used as negative control and a sample well containing 1000ppm solution of antifungal drug (Terbinafine) was used as positive control. The plates were incubated at 28°C for 3 days. The clear zone surrounding each well was indicated its inhibition activity (Bobbarala*et al.,* 2009).

Statistical analysis

The data was subjected to mean, standard deviation and ANOVA by using Statistix 8.1 Program with completely randomized design. All the means were compared by Tukey'sHSD.

Results

Poisoned Food Technique

The results of Poisoned food technique of methanolic spices extracts against *Aspergillus niger*and *A. oryzae* are summarized in Table 2. All the tested plants showed promising antifungal activity against *Aspergillus niger*and *A. oryzae*.

Aspergillus niger

Three concentrations i.e. 25%, 50% and 75% of methanolic extracts were tested. Results were highly significant ($P < 0.05$) and maximum inhibition % age was observed at the concentration of 75% in all the treatments (Table 2). While minimum activity was observed at 25% concentration. The highest inhibition was obtained by *Cuminum cyminum* extract (44.24 ± 2) followed by *Piper nigrum* (43.14 ± 2%), *Myristica fragrans* (Mace) (42.22 ± 2%), *Myristica fragrans* (Nutmeg) (41.48 ± 1.15%), *Carum carvi* (41.48 ± 3.05%), *Brassica juncea* (37.78 ± 2%) and *Curcuma longa* (37.04 ± 2%). The minimum percentage inhibition was observed in *Punica granatum* (19.39 ± 2%), *Cinnamomum zeylenicum* (18.19 ± 4.16%), *Sesamum indicum* (17.48 ± 2%), *Papaver somniferum* (15.55 ± 2%), *Hyoscyamus niger* (15.24 ± 2%) and *Nigella sativa* (13.33 ± 2%). Rest of spices showed least inhibition.

Aspergillus oryzae

The highest inhibition was observed in *Piper nigrum* extract (69.89 ± 2) followed by *Cuminum cyminum* (65.68 ± 3.05%), *Carum carvi* (65.29 ± 1%), *Punica granatum* (62.08 ± 2%), *Myristica fragrans* (Mace) (62.07 ± 2%), and *Myristica fragrans* (Nutmeg) (60.81 ± 1%). Low inhibition was observed in *Mangifera indica* (46.46 ± 2%), *Hyoscyamus*

niger(45.72 ± 1.15%), *Glycyrrhiza glabra* (44.24 ± 2%), *Papaver somniferum* (43.50 ± 3.05%), *Capsicum annuum* (41.85 ± 2.51%) and *Linum usitatissimum* (39.76 ± 2%). Other spices showed moderate percentage inhibition (Table 2).

Agar well diffusion method

Aspergillus niger

All the extracts tested exhibited different zone of inhibition against *Aspergillus niger*. The highest inhibition zone was observed at 75% concentration. All the treatments were highly significant ($P < 0.05$) as indicated in (Table 3). While at 25% concentration, lowest inhibition zone was observed. At 75% concentration, the highest inhibition zone was shown by *Cuminum cyminum* (13.33 ± 1.15), *Myristica fragrans* (Mace) (12.33 ± 0.57), *Curcuma longa* (12.00 ± 2), *Syzygium aromaticum* (12.00 ± 2), *Piper nigrum* (12.00 ± 1), *Zingiber officinale* (12.00 ± 1), *Mentha arvensis* (12.00 ± 1), *Mangifera indica* (12.00 ± 1), *Myristica fragrans* (Nutmeg) (12.00 ± 1), *Cinnamomum tamala* (12.00 ± 1). While the lowest zone of inhibition was shown by *Allium sativum* (9.00 ± 1), *Illicium verum* (9.00 ± 1), *Carum carvi* (9.00 ± 1), *Brassica juncea* (9.00 ± 1), *Amomum subulatum* (8.67 ± 0.57), *Papaver somniferum* (8.33 ± 0.57), *Sesamum indicum* (8.00 ± 1) and *Glycyrrhiza glabra* (7.00 ± 1). Other spices showed moderate zone of inhibition.

Table 1. List of Spices with English name, common name, botanical names and plant parts used.

Sr. no.	English name	Common name	Parts used	Botanical name
1	Carom Seed	Ajwaen	Seed	*Trachyspermum ammi*
2	Turmeric	Haldi	Rhizome	*Curcuma longa*
3	Red Chilli	SurkhMirch	Fruit	*Capsicum annuum*
4	Cumin	SafedZeera	Fruit	*Cuminum cyminum*
5	Coriander	Dhaniya	Leaves	*Coriandrum sativum*
6	Fennel	Saunf	Fruit	*Foeniculum vulgare*
7	Clove	Loang	Flowering bud	*Syzygium aromaticum*
8	Fenugreek	Methi	Leaf	*Trigonella foenum-graecum*
9	Garlic	Lehsan	Bulb	*Allium sativum*
10	Cinnamon	Darchini	Bark	*Cinnamomum zeylenicum*
11	Pomegranate Seed	Anardaana	Seed	*Punica granatum*
12	Anise	Alsi	Seeds	*Linum usitatissimum*
13	Sesame	Til	Seed	*Sesamum indicum*
14	Henbane	Ajwaen Khurasani	Seed	*Hyoscyamus niger*
15	Mace	Javitri	Aril	*Myristica fragrans*
16	Nigella Seed	Kalonji	Seed	*Nigella sativa*
17	Black Pepper	Kali Mirch	Fruit	*Piper nigrum*
18	Liquorice	Mulathi	Stem	*Glycyrrhiza glabra*
19	Emblica Gooseberry	Aamla	Fruit	*Emblica officinalis*
20	Ginger	Adrak	Rhizome	*Zingiber officinale*
21	Mint	Podina	Leaves	*Mentha arvensis*
22	Green Cardamom	ChotiElaichi	Fruit	*Elettaria cardamomum*
23	Black Cardamom	Bari Elaichi	Fruit	*Amomum subulatum*
24	Mango Powder	Amchoor	Fruit	*Mangifera indica*
25	Nutmeg	Jaifal	Seed	*Myristica fragrans*
26	Poppy Seed	Khuskhas	Seed	*Papaver somniferum*
27	Bay Leaf	Tezpatta	Leaf	*Cinnamomum tamala*
28	Star Anise	Baadyaankaphool	Fruit	*Illicium verum*
29	Caraway	Kala Zeera	Fruit	*Carum carvi*
30	Mustard	Sarson	Seed	*Brassica juncea*

Aspergillus oryzae

All the extracts tested exhibited different zone of inhibition against *Aspergillus oryzae*. The highest inhibition zone was observed at 75% concentration. There was significant difference ($P < 0.05$) among treatments as presented in (Table 3). While at 25% concentration, lowest inhibition zone was observed. At 75% concentration, the highest inhibition zone was shown by *Allium sativum* (13.33 ± 0.57), followed by *Cuminum cyminum* (13.00 ± 1), *Trigonella foenum-graecum* (13.00 ± 2), *Trachyspermum ammi* (12.33 ± 1.52), *Myristica fragrans* (Mace) (12.33 ± 0.57), *Piper nigrum* (11.67 ± 1.15) and *Syzygium aromaticum* (11.33 ± 3.05). While the lowest inhibition zone was shown by *Illicium verum* (7.67 ± 0.57), *Carum carvi* (7.33 ± 0.57), *Amomum subulatum* (7.00 ± 1), *Cinnamomum tamala* (7.00 ± 0.57), *Curcuma longa* (6.67 ± 0.57) and *Mentha arvensis* (6.33 ± 0.57).

Table 2. Antifungal activity of spice extracts against *Aspergillus niger* and *Aspergillus oryzae* by poisoned food technique at 75% concentration.

	Inhibition of mycelial growth (%)	
Spices	*A. niger*	*A. oryzae*
Trachyspermum ammi	32.87 ± 10 ^{CDEFG}	51.54 ± 2^{DEFGHIJ}
Curcuma longa	37.04 ± 1.15 ^{ABCDE}	48.89 ± 2 ^{FGHIJKL}
Capsicum annuum	20.54 ± 2.30 ^{HIJK}	41.85 ± 2.51 ^{KL}
Cuminum cyminum	44.24 ± 2 ^{A}	65.68 ± 3.05 ^{AB}
Coriandrum sativum	30.58 ± 2 ^{DEFGH}	48.12 ± 0.57 ^{GHIJKL}
Foeniculum vulgare	21.26 ± 0.57 ^{HIJK}	55.42 ± 0.57 ^{CDEFGH}
Syzygium aromaticum	23.40 ± 3.05 ^{FGHIJK}	57.42 ± 7.21 ^{BCDEFG}
Trigonella foenum-graecum	27.60 ± 3.05 ^{EFGHI}	56.34 ± 1 ^{BCDEFG}
Allium sativum	26.88 ± 5.03 ^{EFGHI}	55.22 ± 2 ^{CDEFGHI}
Cinnamomum zeylenicum	18.19 ± 4.16 ^{IJK}	57.46 ± 2 ^{BCDEFG}
Punica granatum	19.39 ± 2 ^{IJK}	62.08 ± 2 ^{ABC}
Linum usitatissimum	21.62 ± 2 ^{HIJK}	39.76 ± 2 ^{L}
Sesamum indicum	17.48 ± 2 ^{IJK}	48.70 ± 2 ^{FGHIJKL}
Hyoscyamus niger	15.24 ± 2 ^{JK}	45.72 ± 1.15 ^{IJKL}
Myristica fragrans	42.22 ± 2 ^{ABC}	62.07 ± 2 ^{ABC}
Nigella sativa	13.33 ± 2 ^{K}	55.55 ± 2 ^{CDEFGH}
Piper nigrum	43.14 ± 2 ^{AB}	69.89 ± 1 ^{A}
Glycyrrhiza glabra	33.33 ± 2 ^{BCDEF}	44.24 ± 2 ^{JKL}
Emblica officinalis	22.22 ± 1 ^{HIJK}	55.55 ± 1 ^{CDEFGH}
Zingiber officinale	33.07 ± 1 ^{BCDEFG}	57.83 ± 2.08 ^{BCDEF}
Mentha arvensis	33.33 ± 1 ^{BCDEF}	48.89 ± 1 ^{FGHIJKL}
Elettaria cardamomum	26.67 ± 1 ^{FGHI}	57.45 ± 1 ^{BCDEFG}
Amomum subulatum	22.96 ± 2.30 ^{GHIJK}	58.57 ± 1 ^{BCDE}
Mangifera indica	22.22 ± 1 ^{HIJK}	46.46 ± 2 ^{HIJKL}
Myristica fragrans	41.48 ± 1.15 ^{ABC}	60.81 ± 1 ^{ABCD}
Papaver somniferum	15.55 ± 2 ^{JK}	43.50 ± 3.05 ^{JKL}
Cinnamomum tamala	24.44 ± 1 ^{FGHIJ}	54.09 ± 1 ^{CDEFGHI}
Illicium verum	33.33 ± 2 ^{BCDEF}	55.55 ± 2 ^{CDEFGH}
Carum carvi	41.48 ± 3.05 ^{ABC}	65.29 ± 1 ^{AB}
Brassica juncea	37.78 ± 2 ^{ABCD}	50.74 ± 2 ^{EFGHIJK}

*Mean ± Standard Deviation

*Mean values within columns followed by the same letter are not significantly different according to Tukey's HSD test ($P \leq 0.05$).

One way Analysis of Variance was used to determine whether the techniques for antifungal activity differ among each other and also the antifungal activity differs among different type of spices. The analysis shows non-significant difference ($P > 0.05$) among the techniques which shows that there is no difference between two techniques, both the techniques are equally effective. Any of one technique can be used for the determination of antifungal activity. The analysis shows significant difference amongst the spices. Tukey's HSD test shows that *Myristica fragrans* (Mace), *Piper nigrum, Cuminum cyminum, Myristica fragrans* (Nutmeg), and *Trachyspermum ammi* differ significantly from other groups and is statistically significant to each other.

It has been found that leaf extract of *Piper nigrum* inhibits the growth of *Pseudomonas aeruginosa* (Larhsini*et al.,* 2001).

Table 3. Antifungal activity of spice extracts against *Aspergillus niger* and *Aspergillus oryzae* by agar well diffusion method at 75% concentration.

	Mycelial growth (mm)	
Spices	*A. niger*	*A. oryzae*
Trachyspermum ammi	11.00 ± 1 ^{ABCD}	12.33 ± 1.52 ^{AB}
Curcuma longa	12.00 ± 2 ^{ABC}	6.67 ± 0.57 ^{FG}
Capsicum annuum	11 ± 1 ^{ABCD}	10.33 ± 0.57 ^{ABCDE}
Cuminum cyminum	13.33 ± 1.15 ^{A}	13.00 ± 1 ^{A}
Coriandrum sativum	11 ± 2.64 ^{ABCD}	9.33 ± 0.57 ^{BCDEFG}
Foeniculum vulgare	10.67 ± 1.52 ^{ABCD}	10.00 ± 1 ^{ABCDEF}
Syzygium aromaticum	12.00 ± 2 ^{ABC}	11.33 ± 3.05 ^{ABCD}
Trigonella foenum-graecum	11.00 ± 2.64 ^{ABCD}	13.00 ± 2 ^{A}
Allium sativum	9.00 ± 1 ^{BCD}	13.33 ± 0.57 ^{A}
Cinnamomum zeylenicum	9.67 ± 2.08 ^{ABCD}	10.00 ± 1 ^{ABCDEF}
Punica granatum	11.00 ± 1 ^{ABCD}	8.33 ± 0.57 ^{CDEFG}
Linum usitatissimum	9.67 ± 0.57 ^{ABCD}	8.00 ± 1 ^{DEFG}
Sesamum indicum	8.00 ± 1 ^{CD}	9.00 ± 1 ^{BCDEFG}
Hyoscyamus niger	9.33 ± 0.57 ^{ABCD}	9.33 ± 0.57 ^{BCDEFG}
Myristica fragrans	12.33 ± 0.57 ^{AB}	12.33 ± 0.57 ^{AB}
Nigella sativa	10.67 ± 1.15 ^{ABCD}	8.00 ± 1 ^{DEFG}
Piper nigrum	12.00 ± 1 ^{ABC}	11.67 ± 1.15 ^{ABC}
Glycyrrhiza glabra	7.00 ± 1 ^{D}	8.00 ± 1 ^{DEFG}
Emblica officinalis	10.33 ± 0.57 ^{ABCD}	8.00 ± 1 ^{DEFG}
Zingiber officinale	12.00 ± 1 ^{ABC}	8.00 ± 1 ^{DEFG}
Mentha arvensis	12.00 ± 1 ^{ABC}	6.33 ± 0.57 ^{G}
Elettaria cardamomum	9.33 ± 0.57 ^{ABCD}	8.67 ± 0.57 ^{CDEFG}
Amomum subulatum	8.67 ± 0.57 ^{BCD}	7.00 ± 1 ^{EFG}
Mangifera indica	12.00 ± 1 ^{ABC}	10.00 ± 1^{ABCDEF}
Myristica fragrans	12.00 ± 1 ^{ABC}	8.67 ± 0.57 ^{CDEFG}
Papaver somniferum	8.33 ± 0.57 ^{BCD}	8.00 ± 1 ^{DEFG}
Cinnamomum tamala	12.00 ± 1 ^{ABC}	7.00 ± 0.57 ^{EFG}
Illicium verum	9.00 ± 1 ^{BCD}	7.67 ± 0.57 ^{EFG}
Carum carvi	9.00 ± 1 ^{BCD}	7.33 ± 0.57 ^{EFG}
Brassica juncea	9.00 ± 1 ^{BCD}	8.00 ± 1 ^{DEFG}

*Mean ± Standard Deviation

*Mean values within columns followed by the same letter are not significantly different according to Tukey's HSD test ($P \leq 0.05$).

Discussion

Accordingly, in the present study, efficacy of 30 spice extracts was evaluated against two fungal species i.e. *Aspergillus niger* and *A. oryzae* at 25, 50 and 75% concentrations in methanol. The extracts were subjected to antifungal assays *viz.* Poisoned food technique and Agar well diffusion method against both the fungal pathogens.

Present study revealed highest inhibition activity by *Myristica fragrans* (Mace) spice extracts with 12.33mm zone of inhibition against *Aspergillus niger* and *Aspergillus oryzae*. Similar studies were carried out by Pooja*et al.*,(2012) in which *Myristica fragrans* (Mace) spice extracts showed maximum antimicrobial activity with 17mm and 19mm zone of inhibition for methanolic extract against *Candida albicans* and *Aspergillus niger*respectively.

Present study exhibited that nutmeg have strong antifungal activity against *Aspergillus niger* and *Aspergillus oryzae* at 75% concentration.

The findings of Gupta *et al.*,(2013) supports our results. According to their findings, nutmeg seeds have strong antimicrobial activity against important pathogenic bacteria and fungi. Another study of Cho *et al.,* (2007) reported compounds which have antifungal activity isolated from methanolic extract of nutmeg i.e. Three lignans, mesodihydroguaiareticacid, erythro austrobailignan-6 and nectandrin-B.

In present study, *Piper nigrum* and *Trachyspermum ammi* inhibited 43.14% and 32.87% *Aspergillus niger*which are in agreement with the findings of Avasthi *et al.,* (2010). According to their findings, *Trachyspermum ammi* and *Piper nigrum* (48.93%, and 46.2%) showed inhibition against *Aspergillus niger*.

Trachyspermum ammi in our studyshowed 32.87% and 51.54% inhibition against *Aspergillus niger* and *Aspergillus oryzae* respectively. Similar investigations were evaluated by Murthy *et al.*,(2009) by finding that Ajwaen essential oil was active *against Aspergillus niger, Aspergillus oryzae, Aspergillus ochraceus, Aspergillus flavus, Fusarium moniliforme, and Penicillium* sp. In another study, it has been found that the essentialoil of ajwaen inhibited growth of *Aspergillus niger*(Tiwari*et al.,* 2003).

Cuminum cyminum in present study showed 44.24% and 65.68% inhibition against *Aspergillus niger* and *Aspergillus oryzae* at 75% concentration. Similar studies were carried out about cumin oil for their antifungal activity by El-Said and Goder(2014) who evaluated antifungal activity of cumin essential oils on mycelia growth of 90 isolates of fungi.

Their results showed that cumin oil was highly effective against all the isolates of tested fungi. It completely inhibited mycelial growth of all fungi at a concentration of 100%. While in our study, highest inhibition of mycelial growth of *Aspergillus niger* and *Aspergillus oryzae* were recorded at a concentration of 75%. In another study, it has been found that cumin oils showed complete fungal inhibition against *Penicillium italicum* at concentration of 24 and 48 µl/ml, respectively (Anjum and Nosheen, 2012). According to the study of Kamble (2015), Cumin seed oil showed strong inhibition with growth inhibition zonesranging from 27 to 72 mm against all clinical isolates of *C. albicans* and non-albicans *Candida*. While in present study, *Cuminum cyminum* showed 13.33 and 13 mm growth inhibition zone against *Aspergillus niger* and *Aspergillus oryzae* respectively, in agar well diffusion method.

Conclusion

All the spice extracts showed significant inhibition activity against both the tested fungal pathogens *Aspergillus niger* and *A. oryzae*. *Aspergillus niger* is more aggressive and resistant fungus as compare to *Aspergillus oryzae*. Furthermore, some of the spices extract such as *Cuminum cyminum*, *Myristica fragrans*, *Piper nigrum* and *Curcuma longa* possessed the highest inhibitory activity which can be utilized for drug discovery.

References

Anjum T, Akhtar N. 2012. Antifungal Activity of Essential Oils Extracted From Clove, Cumin and Cinnamon Against Blue Mold Disease on Citrus Fruit. International Conference on Applied Life Sciences. Turkey, September 10-12.

Aktug SE, Karapinar M. 1986. Sensitivity of some common food-poising bacteria to thyme, mint and bay leaves. International Journal of FoodMicrobiology**3,** 349-354.

Avasthi S, Gautam AK, Bhadauria R. 2010. Antifungal activity of plant products against *Aspergillus niger*: A potential application in the control of a spoilage fungus. An International Journal **2,** 53-55.

Baser KHC, Kurkcuoglu M, Ozek T. 1992.Composition of the Turkish cumin seed oil.Journal of Essential Oil Research**4,** 133-138.

Bedin C, Gutkoski SB, Wiest JM. 1999.Atividadeantimicrobiana das especiarias. HigieneAlimentar **13,** 26-29.

Bobbarala V, Katikala PK, Naidu KC, Penumajji S. 2009. Antifungal Activity of Selected Plant Extracts against Phytopathogenic Fungi *Aspergillus niger* F2723. Indian Journal of Science and Technology **2,** 87-90.

Borges P, Pino J. 1993. The isolation of volatile oil from cumin seeds by steam distillation. Nahrung**2,**123-126.

Bugno A, Almodovara AB, Pereira TC, Pinto TA, Sabino M. 2006. Occurrence of toxigenic fungi in herbal drugs. Brazilian Journal of Microbiology **37,** 1-8.

Cho JY, Choi GJ, Son SW, Jang KS, Lim HK, Lee SO, Sung ND, Cho KY, Kim JC. 2007. Isolation *and*antifungal activity of lignans from *Myristica fragrans* against various plant pathogenic fungi. Pest Management Science **63,** 935–940.

El-Said AHM, Goder EH. 2014. Antifungal Activities of *Cuminum cyminum* and *Pimpinella anisum* Essential Oils. International Journal of Current Microbiology and Applied Sciences **3,** 937-944.

Gautam AK, Bhadauria R. 2009.Detection of toxigenic fungi and mycotoxins in medicinally important powdered herbal drugs.The International Journal of Microbiology **7,** 182-187.

Gautam AK, Bhadauria R. 2008.Occurrence of Toxigenic Moulds and Mycotoxins in Ayurvedic Medicine Trifla Churn.Journal of Mycology and Plant Pathology **3,** 664-666.

Germano PML, Germano MIS. 1998.Importancia e riscos das especiarias. HigieneAlimentar**12,** 23-31.

Gupta AD, Bansal VK, Babu V, Maithil N. 2013. Chemistry, antioxidant and antimicrobial potential of nutmeg (Myristica fragrans Houtt). Journal of Genetic Engineering and Biotechnology **11,** 25–31.

Ibrahim MB. 1997. Anti-microbial effects of extract leaf, stem and root bark of *Anogeissusleiocarpus* on *Staphylococcus aureaus*, *Streptococcus pyogenes*, *Escherichia coli* and *Proteus vulgaris*. International Journal of Pharmaceutical Research and Development **2,** 20-30.

Kamble VA. 2015. *In vitro* Anti-Fungal Activity of *Cuminum cyminum* (Cumin Seed) Essential Oil againstClinical Isolates of *Candida* Species. American Journal of Phytomedicine and Clinical Therapeutics.**3,** 264-275.

Kapoor A. 1997.Antifungal activity of fresh juice and aqueous extracts of turmeric and ginger.Journal of Physiological Research **10,** 59-62.

Kizil S, Sogut T. 2003.Investigation of antibacterial effects of spices.Crop Research **3,** 86-90.

Larhsini, ML, Oumoulid HB, Larze M, Bousaid K, Bekkouche Jana M. 2001.Antimicrobial activity of some Moroccan Medicinal Plants.PhytotherapyReserach **15,** 250-252.

Legan JD, Voysey PA. 1991. Yeast spoilage of bakery products and ingredients. Journal of AppliedBacteriology**70,** 361-371.

Mahesh B, Satish S. 2008.Antimicrobial activity of some important medicinal plant against plant and human pathogens.World Journal of Agricultural Sciences **4,** 839-843.

Mann A, Banso A, Clifford LC. 2008. An antifungal property of crude plant extracts from *Anogeissusleiocarpus* and *Terminaliaavicennioides*. Tanzania Journal of Health Research **10,** 34-38.

Murthy PS, Borse BB, Khanum H, Srinivas P. 2009. Inhibitory effects of Ajowan (*Trachyspermum ammi*) ethanolic extract on *A. ochraceus*growth and ochratoxin production. Turkish Journal of Biology**33,** 211-217.
http://dx.doi.org/10.3906/biy-0805-13.

Nwaopara A, Anibeze C, Akpuaka F, Nwaopara S. 2009. Antimicrobial Potentials of Yaji-Spices: The Constituents of a Complex Nigerian Suya Meat Sauce Inducing Histological Investigations. The International Journal of Alternative Medicine**6**.

Ogundipe O, Akinbiyi O, Moody JO. 1998. Antibacterial activities of essential ornamental plants. Nigeria J Natural Products & Medicine **2,** 46-47.

Penkhae W, Chaungwanit P,Poovarodom N, Nitisinprasert S. 2005.In vitro Antifungal Activity and Spice Extracts against Food Spoilage Fungi.Kasetsart Journal-Natural Science **39,** 400-405.

Pooja V, Sanwal H, Goyal A, Bhatnagar S, Srivastava AK. 2012. Activity of *Myristica Fragrans* and its effect against Filamentous and Non-FilamentousFungus. International Journal of Pharmacy and Pharmaceutical Sciences **4,** 538-540.

Rajani P, Sridevi V, Lakshmi MVVC, Kumari SPK. 2012. Inhibitory Effect of Aqueous Plant Extracts on the Growth of Aflatoxin Producing *Aspergillus Parasiticus* (NCIM 898). International Journal of Engineering Science and Advanced Technology **2,** 365-371.

Ristori CAM, Pereira S, Gelli DS O. 2002.efeito da pimenta do reinomoídafrente a contaminacaoin vitro com *Salmonella rubislaw*. Revista do Instituto Adolfo Lutz **61,** 131-133.

Shelef LA. 1983. Antimicrobial effects of spices. Journal of Food Safety,6, 29-44.

Skrinjar MM, Nemet NT, 2009. Antimicrobial effects of spices and herbs essential oils. APTEFF **40,** 195-209.

Tiwari TN, Chansouria JPN, Dubey NK. 2003. Antimycotic potency of some essential oils in the treatment of induced dermatomycosis of an experimental animal. Pharmaceutical Biology **41,** 351-356.
http://dx.doi.org/10.1076/phbi.41.5.351.15935